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THE WEAKLY BASIC ALKALOIDS OF LYCOPODIUM LUCIDULUM

by

©

D. S. NKUNIKA

A THESIS
SUBMITTED TO THE FACULTY OF GRADUATE STUDIES
IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR
THE DEGREE OF DOCTOR OF PHILOSOPHY

DEPARTMENT OF CHEMISTRY
EDMONTON, ALBERTA
DECEMBER, 1967

UNIVERSITY OF ALBERTA

FACULTY OF GRADUATE STUDIES

The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies for acceptance, a thesis entitled,

THE WEAKLY BASIC ALKALOIDS OF LYCOPODIUM LUCIDULUM, submitted by Dalson Simbi NKUNIKA, in partial fulfilment of the requirements for the degree of Doctor of Philosophy.

ACKNOWLEDGEMENTS

The author expresses his thanks to the following:

Mr. M. A. Stilwell who collected and identified the plant material.

Mr. J. McCutcheon who isolated the crude alkaloids.

Mr. R. N. Swindlehurst and his associates who recorded the infrared, optical rotatory and nuclear magnetic resonance spectra.

Mr. Glen Bigam, Peter Gibbs and their associates who obtained the HA-100 or HR-100 nuclear magnetic resonance spectra and performed the irradiation experiments.

Dr. A. N. Hogg, Mr. A. I. Budd and their associates who obtained most of the mass spectra.

The Ministry of Education, Republic of Zambia, who provided a generous maintenance grant during the last two years, and the Department of Chemistry of the University of Alberta for the award of a teaching assistantship.

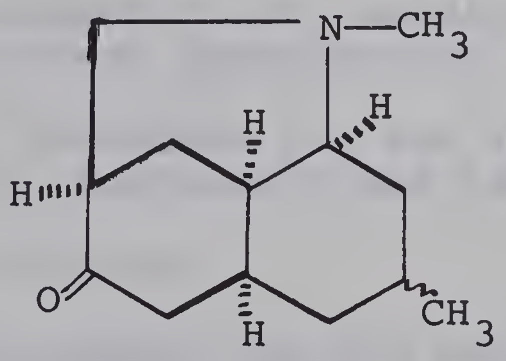
Dr. W. A. Ayer, without whose guidance and knowledge this work would not have been possible.

My wife, Ruth, for her great patience and encouragement.

ABSTRACT

The weakly basic alkaloids of Lycopodium lucidulum Michx have been investigated. A method for the separation of the weak bases utilizing a combination of counter-current distribution and dry-column chromatography (48) has been developed. The isolation of four new alkaloids, luciduline, lucidine-A, lucidine-B and lycolucine is reported. Spectral and chemical degradative evidence shows that luciduline has the structure and stereochemistry shown by structure A.

Chemical dehydrogenation, spectral and chemical data indicate that lucidine-A, lucidine-B and lycolucine are hexacyclic alkaloids which possess both a 2,3,6-trisubstituted pyridine system and a 7-methylquinoline system.



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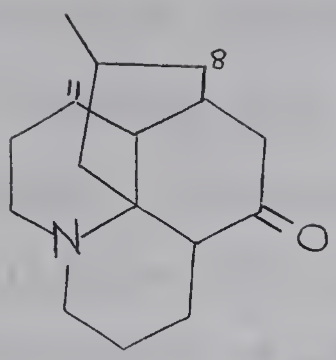
INTRODUCTION

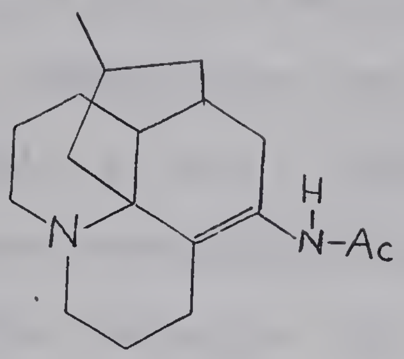
The presence of alkaloids in the genus Lycopodium was first reported by Bödeker in 1881 when he reported the isolation of an alkaloid from *Lycopodium complanatum* (1). Some sixty years later Marion and Manske undertook a systematic investigation of Lycopodium species. They investigated about a dozen species and isolated and characterized some thirty different alkaloids (2). It was, however, a further decade before the first structure of a Lycopodium alkaloid, annotinine, was elucidated by Wiesner, Valenta and co-workers in 1956 (3). This structure of annotinine was confirmed by X-ray crystallography (4). In 1960 MacLean and Harrison (5) reported the structure of lycopodine, and since then the structures of well over thirty alkaloids have been elucidated.

The known alkaloids of lycopods can be divided into five structural types. The first group, the lycopodine type, includes the largest number of the alkaloids. Lycopodine (I), clavolonine (6), lycoclavine (7), L20 (8), lycofawcine (9), lycodoline (10), lycofoline (11), lofoline (6,12), annofoline (13,14), acrifoline (15), L23 (16) and flabelline (17) belong to this class.Flabelline (II), unlike the other lycopodine type of alkaloids, is dinitrogenous but it has the same skeleton as lycopodine.

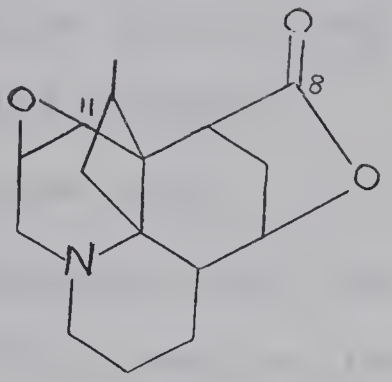
The second group of bases, the annotinine type, comprises the three alkaloids annotinine (4), annotine (18) and lycorotine (19). These alkaloids occur in *L. annotinum*, botanically



I



II



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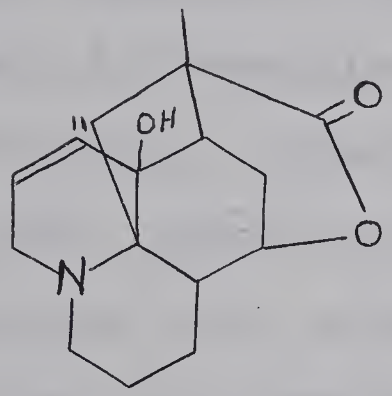
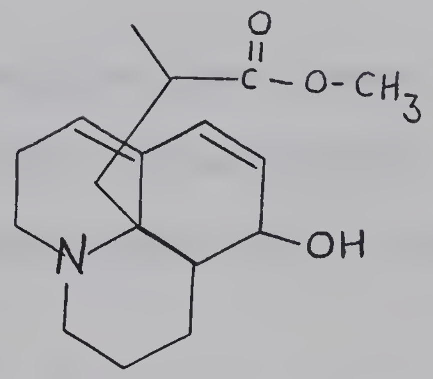
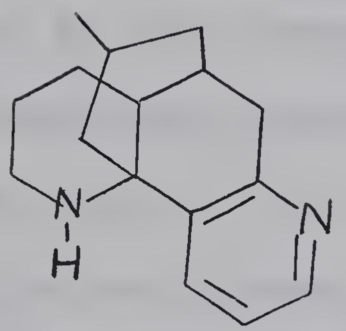


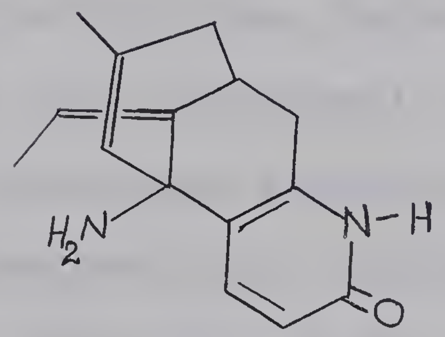
IV



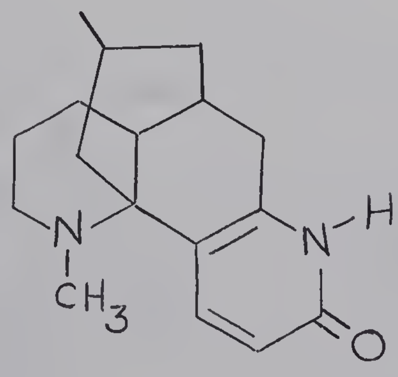
V



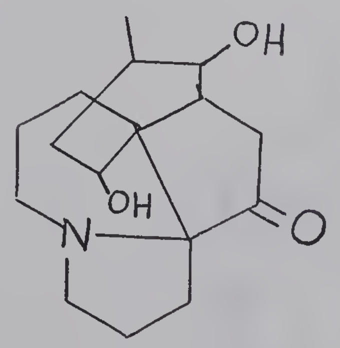
VI



VII



VIII



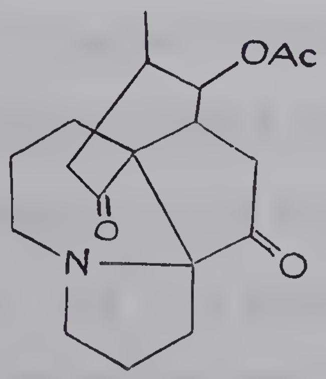
IX

the most primitive of the series, and possess similar levels of oxidation patterns at C8 and C11 as shown in III, IV and V.

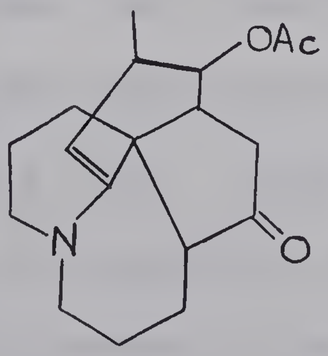
Lycodine (20) (VI), exemplifies the third group of alkaloids. Sauroxine (26), α- and β- obscurine (21), flabellidine (22), and des-N-methylhydroxyobscurine (22) are lycodine-type of alkaloids. Selagine (23) (VII), has one less carbon atom than the typical lycodine class, but it is appropriately included in this group since its structure is similar to that of β-obscurine (VIII).

Recently, Inubushi and co-workers isolated two known alkaloids, lycodoline and lycodine and four new alkaloids, serratinine (24), serratanine (24), serratinidine (25), and serratine (27) from L. serratum.

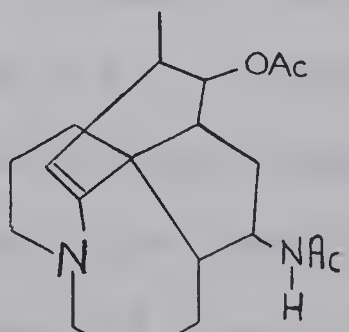
As a result of extensive degradative work (24), they arrived at the structure IX for serratinine. Reduction of dehydromonoacetylserratinine (X) with zinc-acetic acid yielded some of the skeletally rearranged anhydrocompound XI, which was converted to the compound XII. Acetylserratinidine was found to be identical with compound XII.



X



XI



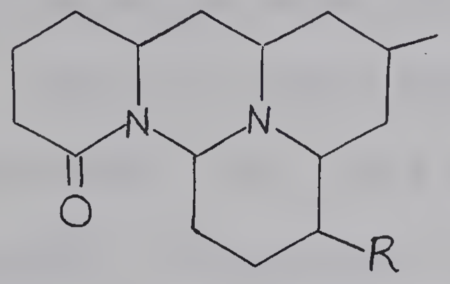
XII

The structure of serratine (27) was established in a similar way. Serratinine has been transformed into fawcettidine (28), isolated from L. fawcettii (30), thus demonstrating the presence of serratinine type of alkaloids in a species other than L. serratum. In a coöperative effort between three groups (29), it has been shown that fawcettimine (Base A), also from L. fawcettii (30), belongs to this group of alkaloids. So far this fourth group comprises fawcettimine, fawcettidine, serratinine, serratinidine and serratine.

The fifth type of alkaloids has come to light as a result of the re-investigation of the alkaloids of L.cernuum. L.cernuum is unique among Lycopodium species in that it does not contain any of the common lycopodine or lycodine type of alkaloids. L. cernuum contains the alkaloids cernuine and lycocernuine and small amounts of dihydrodeoxycernuine (31, 32). The structures of cernuine (XIII) and lycocernuine (XIV) have been proposed largely on the basis of the isolation of 2-n-butyl-4-methyl-6-pentylpyridine (XV) from the dehydrogenation of cernuine, biosynthetic considerations, and other degradative results.

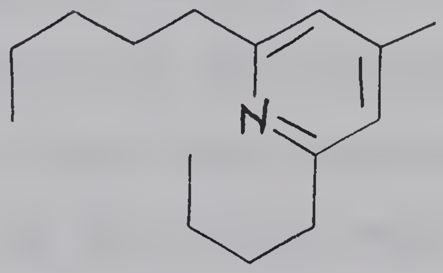
Conroy suggested that the alkaloids of lycopods could be biosynthesized through the cyclization of two 3,5,7 - triketo-octanoic acid equivalents (33). Ayer and co-workers assumed that the C-4 methyl of the pyridine represents the C-methyl of cernuine and that this methyl group is derived from the terminal methyl of one of the 3,5,7 - triketo-octanoic acid

chains. Aldol condensation of its adjacent carbonyl with the C-methyl of the other poly-β-ketoacid chain followed by condensation with the two moles of ammonia led to a structure (XIII) for cernuine. The structure so derived has since been confirmed

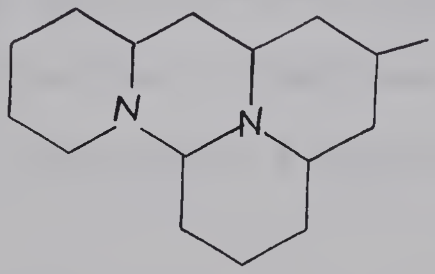


XIII R = H

XIV R = OH



XV



XVI

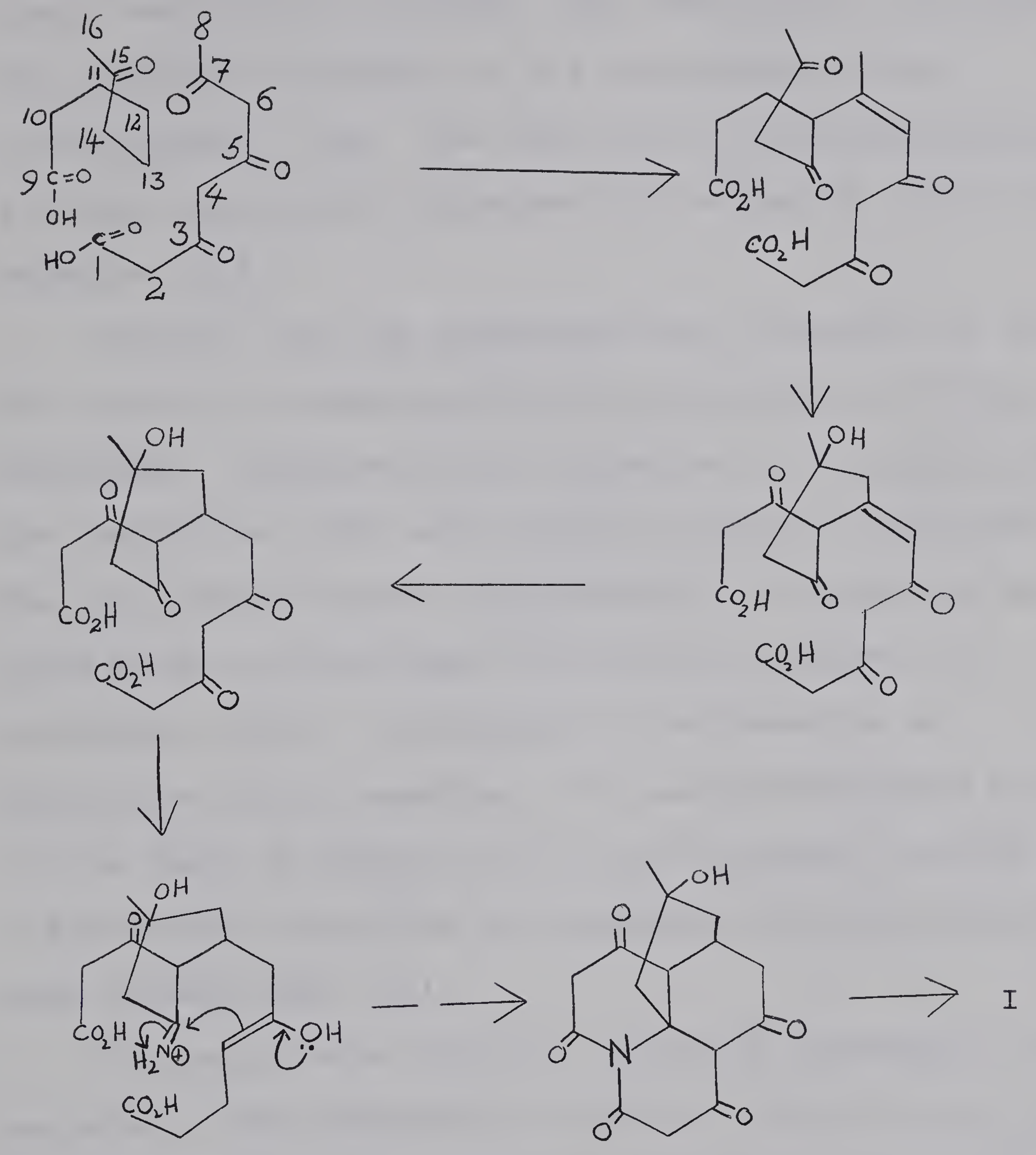
by further chemical degradation and also by the total synthesis of a compound (34), dihydrodeoxyepiallocernuine (XVI), with the same skeleton as cernuine. The biogenetic significance of these results will be discussed again in connection with the biosynthesis of these alkaloids.

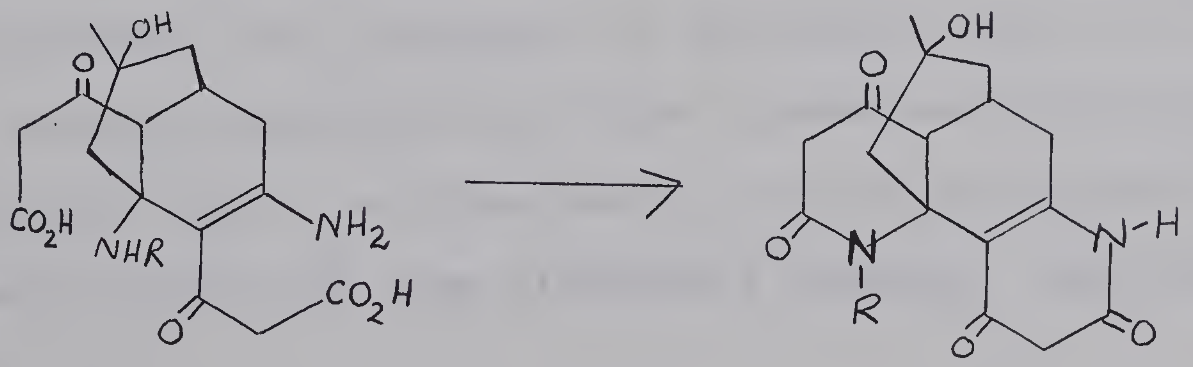
Biogenetic considerations have played an important role in the elucidation of the Lycopodium alkaloids, notably, cernuine and lycocernuine (31,32), lycodine (20), annotine (18) and more recently fawcettidine (28), and fawcettimine (29). In view of the usefulness of biogenetic considerations in structure determination, the biogenesis of the five classes of lycopodium alkaloids is discussed in some detail.

Shortly after Wiesner's elucidation of the structure of annotinine (35), and more generally, after the structures of selagine, lycopodine, lycodine and the obscurines were known, an ingenious, and indeed very fruitful suggestion was made concerning the biosynthesis of annotinine and other lycopodium alkaloids (33). As already mentioned, the biogenesis is explained by the condensation of two 3,5,7 - triketo-octanoic acid equivalents with one or more equivalents of ammonia. Although the order of stops is unknown, the sequence shown in the chart on page 7 has been suggested for the biosynthesis of lycopodine.

The recent discovery (31) of Lycopodium alkaloids in which it appears that the only carbon-carbon bond formed between the two polyketoacid chains is the C-8 to C-15 bond strongly indicates the possibility that the first aldol condensation occurs between the C-8 methyl of one chain and the C-15 carbonyl of the other to form XVII. This would then be a common intermediate in the formation of all lycopodium alkaloids.

Similarly, the lycodine type of alkaloids can be derived via the intermediacy of XVIII.


OH
HO
CO2H
H2N⊕
I


OH
CO2H
NHR
NH2
N-H
N
R

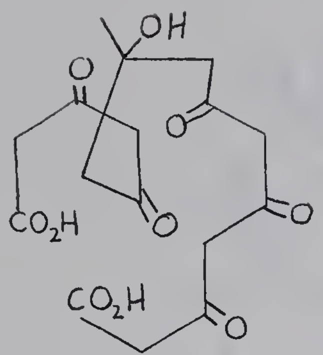
XVIII

XIX

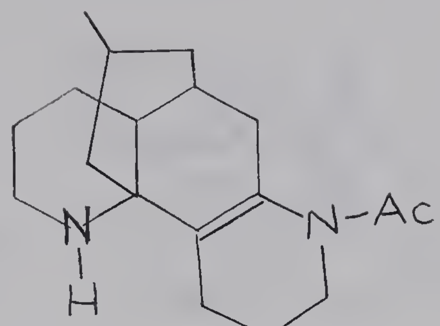
Lactamization of XVIII leads to XIX and unexceptional steps lead to the pyridones (the obscurines, and sauroxine), the pyridine (lycodine) or the tetrahydropyridine (flabellidine - XX). The loss of C-9 by decarboxylation followed by suitable adjustment of oxidation levels forms selagine (VII).

Wiesner (35) has suggested that lycopodine is possibly the central intermediate in the biosynthesis of these alkaloids. Oxygenation of lycopodine at C-8 could give the annofoline (XXI) and acrifoline type of alkaloids. The similarity between the hemiketal of annofoline and annotinine is significant in that the hemiketal of acrifoline (XXII) could lead to the formation of annotinine (III), annotine (IV) and lyconnotine(V) as shown in the chart on pages 9-10. It is of interest to note that a laboratory conversion of lycopodine into annofoline has been accomplished (36).

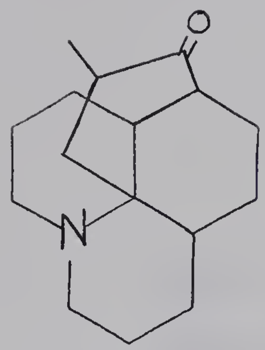
The serratinine group of alkaloids possesses a novel skeleton. The presence of lycodoline (XXIII) in L. serratum led Inubushi and co-workers to speculate that (37) serratinine is probably biosynthesised from lycodoline in the plant. Serratine (XXLV) is structurally similar to serratinine and is formed through the same biogenetic pathway. (See chart on page 12.


OH
O
O
O
O
O
CO_2H
CO_2H

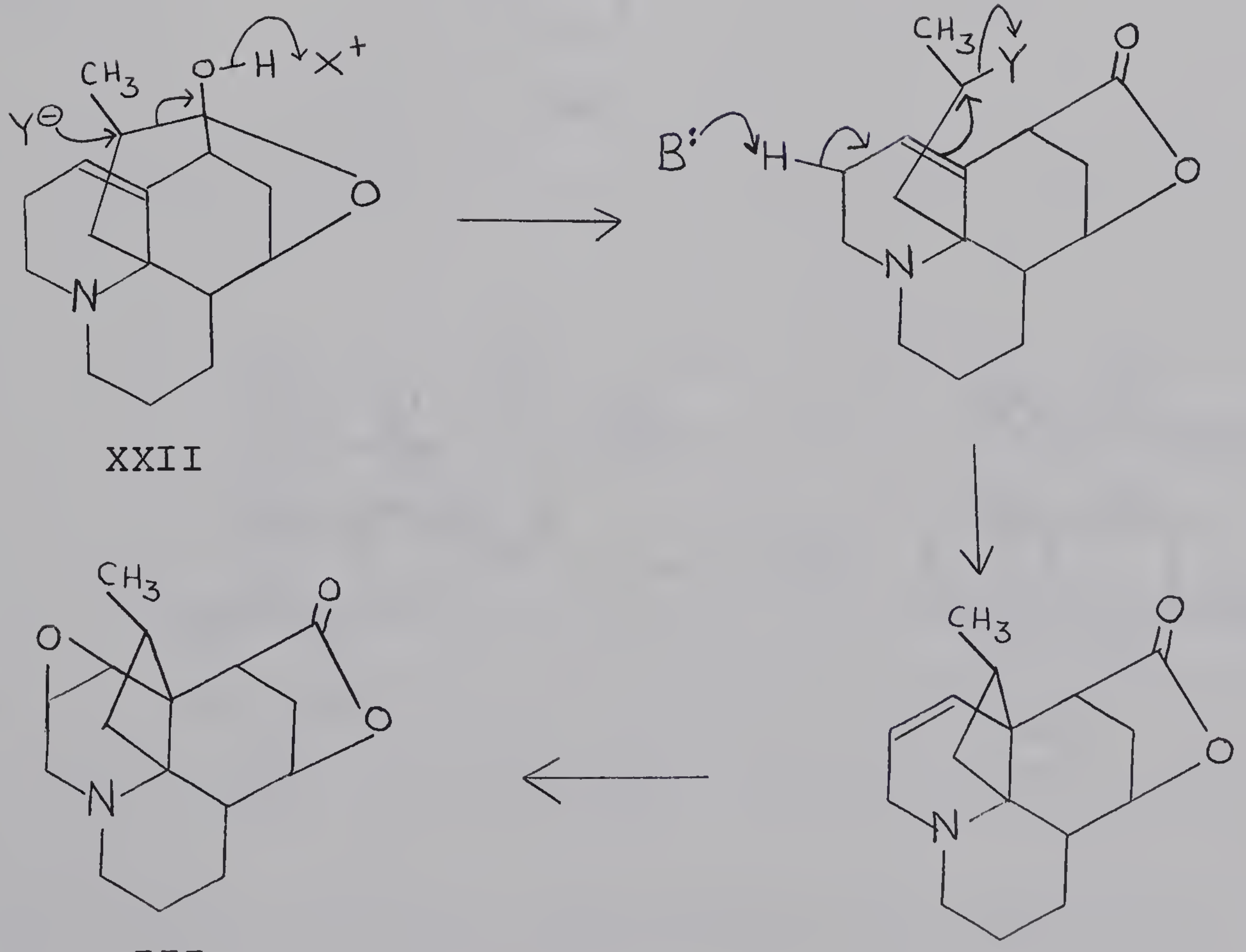
XVII



XX

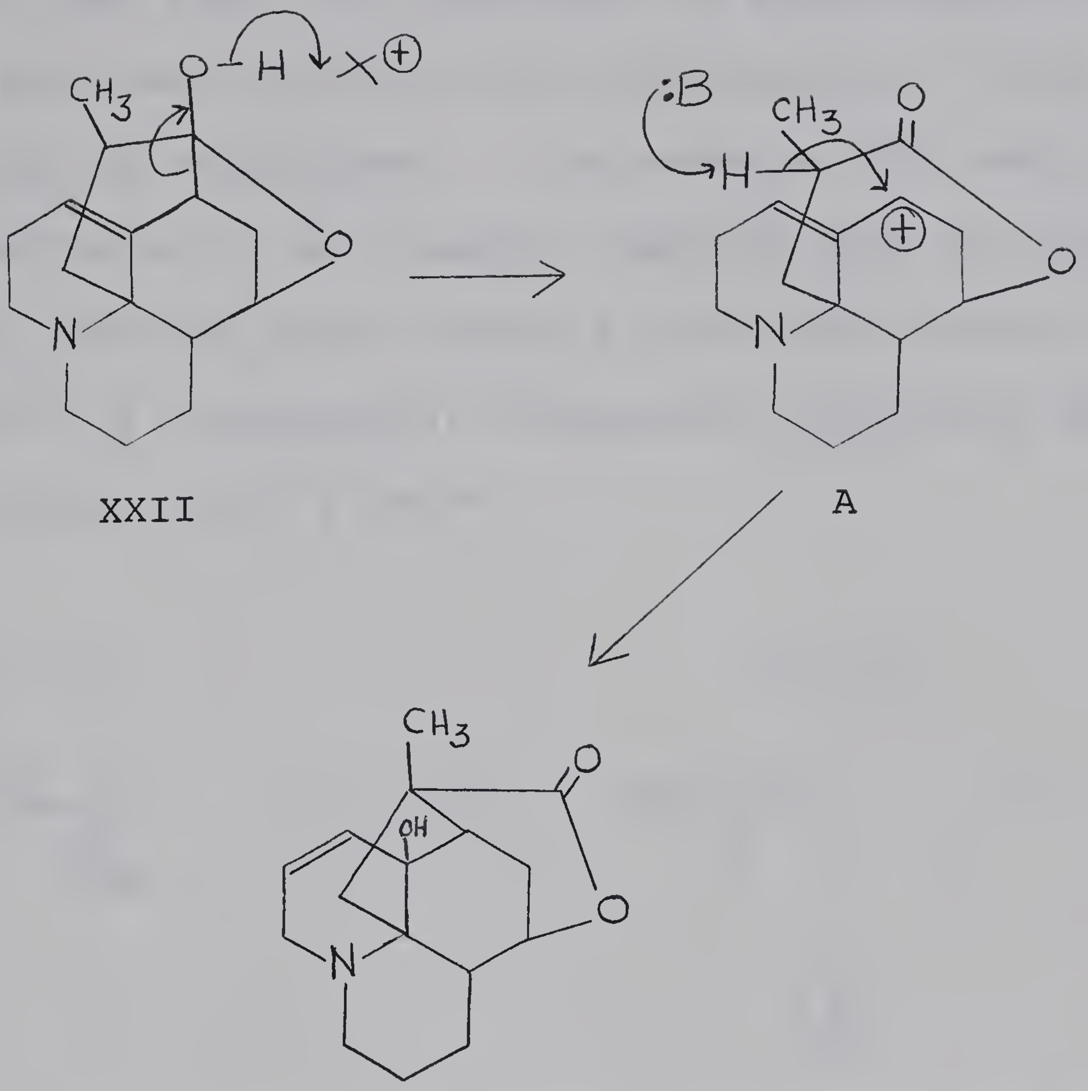


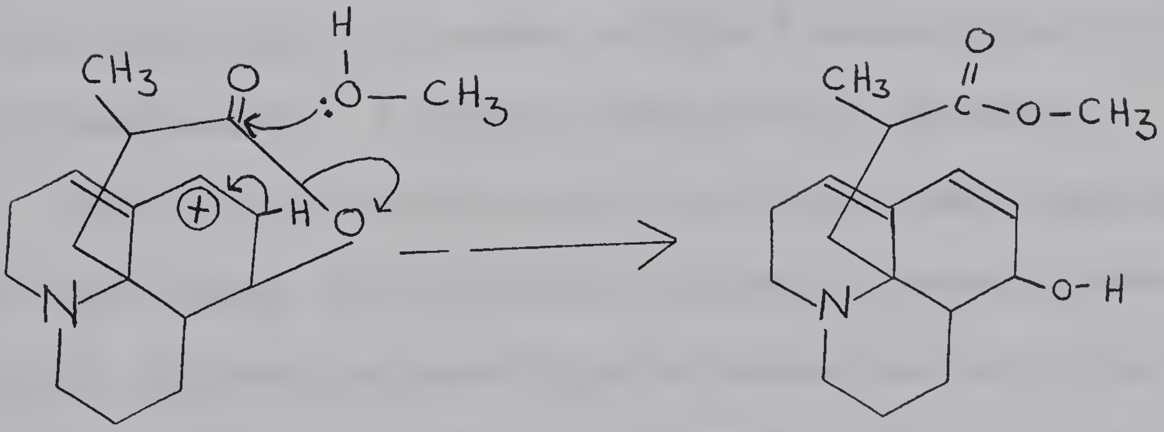
XXI


CH_3
$Y^\ominus$
O-H
X^+
O
N
B:
H
CH_3
Y
O
O
N
CH_3
O
O
N
CH_3
O
O
O
N

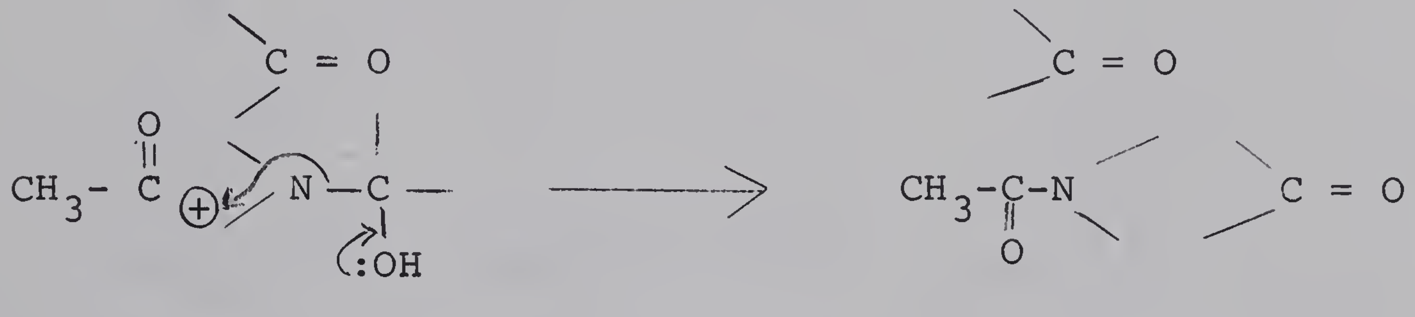
XXII

III


CH3
O-H
X⊕
O
N
XXII
:B
CH3
O
H
⊕
O
N
A
CH3
O
OH
O
N
IV


CH3
O
H
:O-CH3
⊗
H
O
N
A
CH3
O
C-O-CH3
O-H
N
V

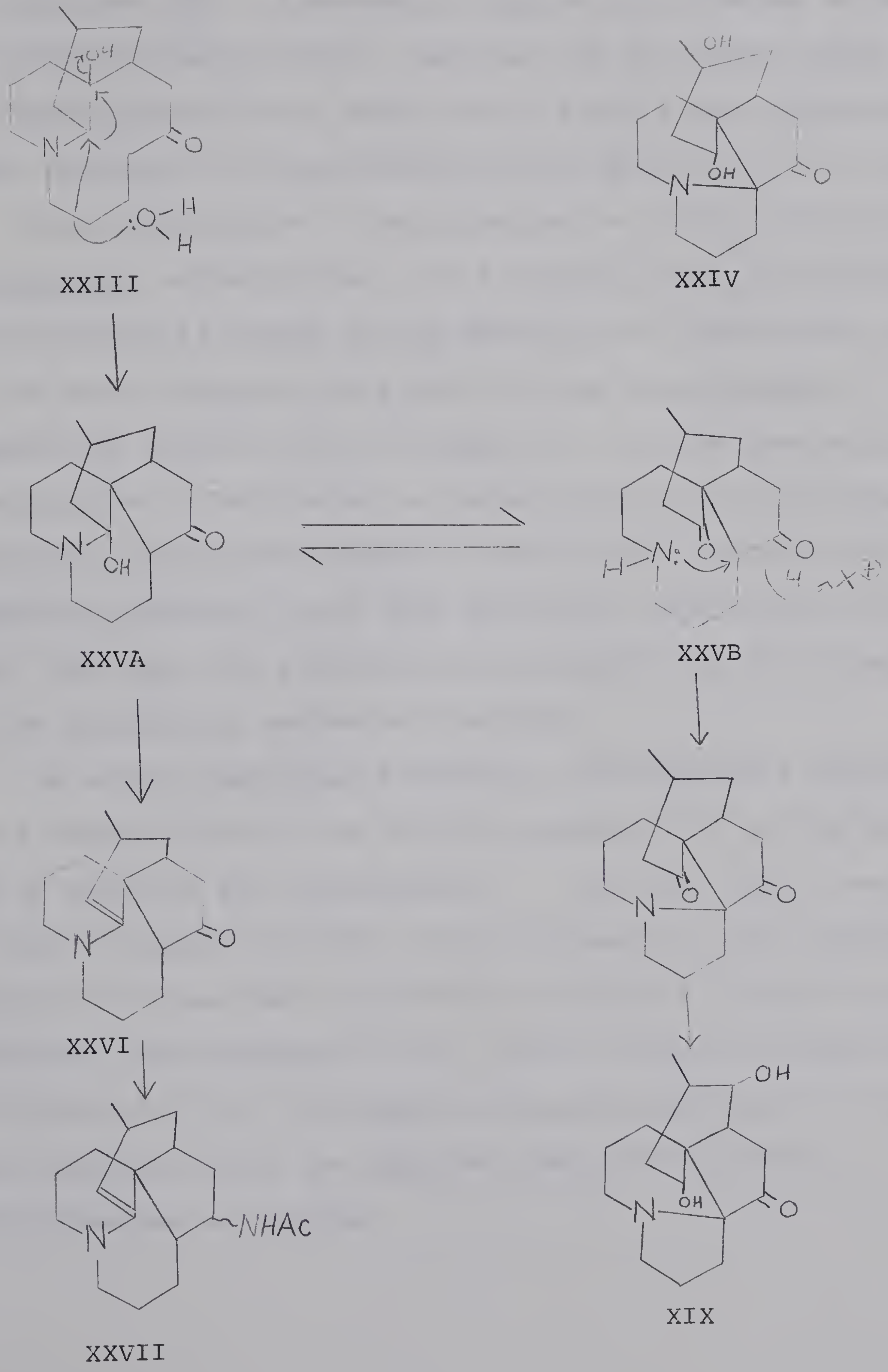
For some time (38), the reactions of fawcettimine and fawcettidine have been difficult to rationalize in terms of lycopodine type of structures. For example, the acetylation of fawcettimine to an N-acetyl compound with no hydroxyl or amino group and one extra carbonyl group was rationalized only in terms of a transannular interaction involving the partial structures A and B below:



A B

Shortly after the recent publication of the structure of serratinidine and fawcettidine it was thought likely that fawcettimine might have the structure XXVA. The acetylation product is then explicable in terms of the transannular interaction already mentioned. A direct correlation between fawcettimine (Base A) and serratinine (IX) has been reported (29). It has been shown that dihydro-N-acetyl-fawcettimine is identical with N-acetyl-chanodihydro-8-deoxyserratinine (XXIX). The series of conversions are summarized in Chart I on page 15 .

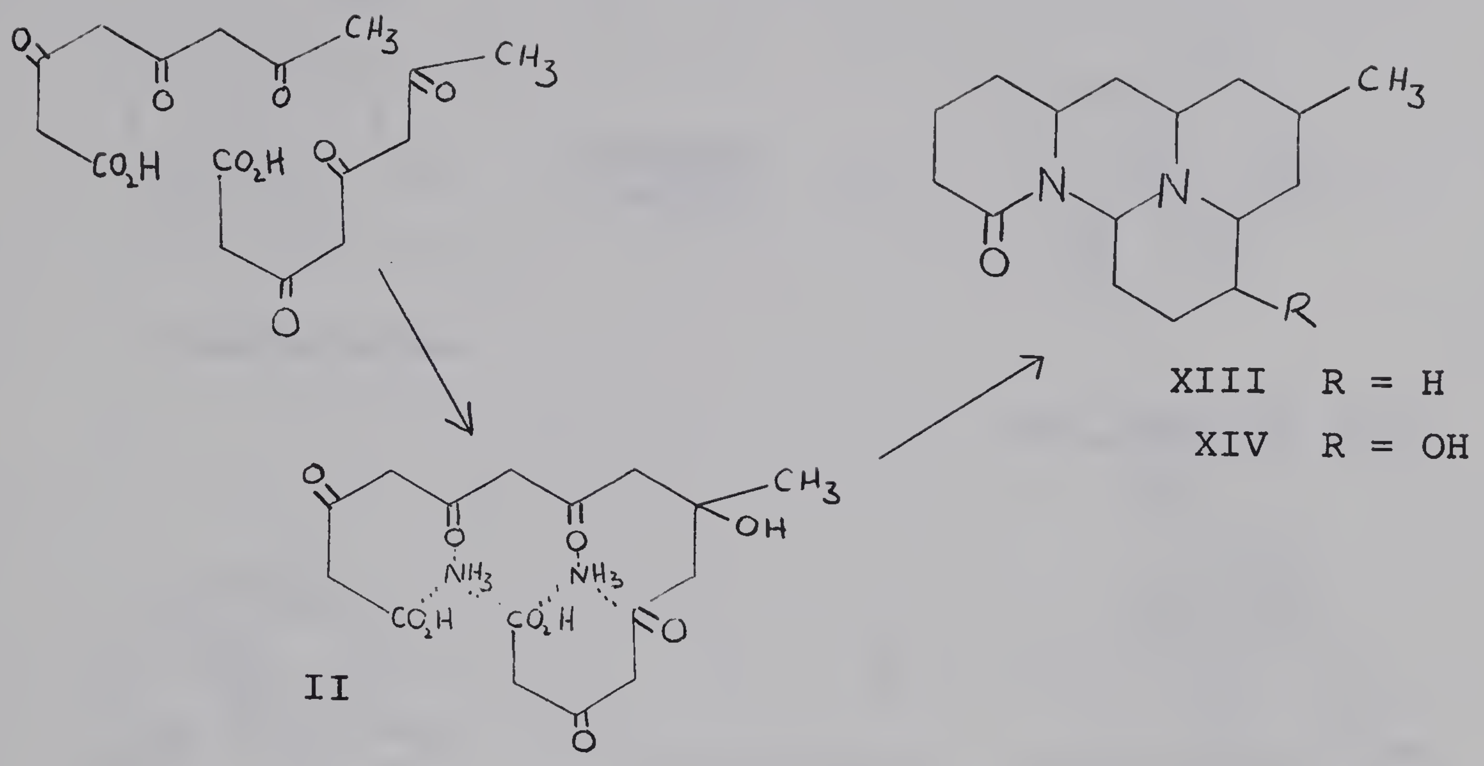
Serratinine has been transformed into fawcettidine (25). Serratinine was converted into monoacetylserratinine which was oxidized to the diketone XXX. Desulfurisation of the ethylene-


OH
O
N
:O–H
H
XXIII
OH
OH
N
O
XXIV
N
OH
O
XXVA
H–N:
O
O
H
X⊕
XXVB
N
O
XXVI
N
O
O
N
NHAc
XXVII
OH
N
OH
O
XIX

thioketal of XXX, followed by hydrolysis and oxidation, gave the diketone XXXI. Zinc-acetic acid on the diketone afforded the dehydro-compound XXXII. Reduction of the ketone XXXII gives the dehydroaminoalcohol XXXIII which has the same properties as those reported for dihydrofawcettidine.(See Chart II on page 15).

Thus fawcettidine is very similar to a minor alkaloid of L. serratum, serratinidine. It is possible that in the plant serratinidine is formed by the amination of fawcettidine and is, in turn, biogenetically derived from fawcettidine by dehydration (See the Chart on page 12). In the laboratory, fawcettimine is dehydrated to fawcettidine by the pyridine-phosphorus oxychloride method. The natural occurrence of compounds possessing what were previously hypothetical structures XXVA, XXVB and XXVI provides strong support for the biogenesis of the serratinine series of alkaloids.

As already mentioned biogenetic considerations constituted a crucial step in the initial proposal (31) of the structure of cernuine and lycocernuine. The fact that a wealth of other evidence including total synthesis of the cernuine skeleton (34) has been successfully adduced to support this structure lends credence to the likely polyketide origin of the alkaloids of all the known Lycopodium alkaloids. It has been postulated that the cernuine group of alkaloids is biosynthesized as follows:



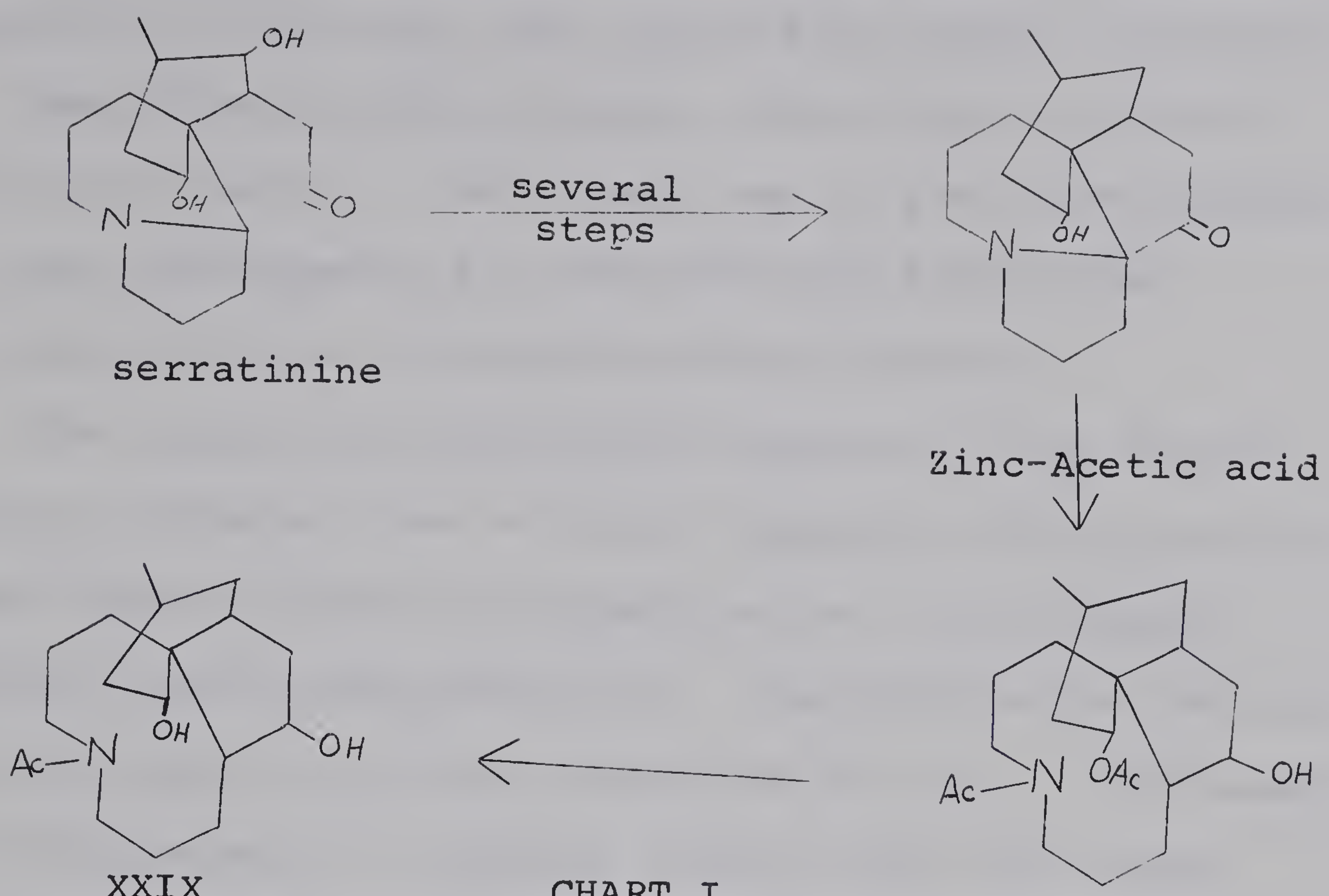
II

XIII R = H

XIV R = OH

Structure II or its biological equivalent could, quite possibly, represent the first condensation product in all the alkaloids. The formation of the particular group or groups of alkaloids probably depends on the geometrical requirements of the enzyme systems in the individual species.

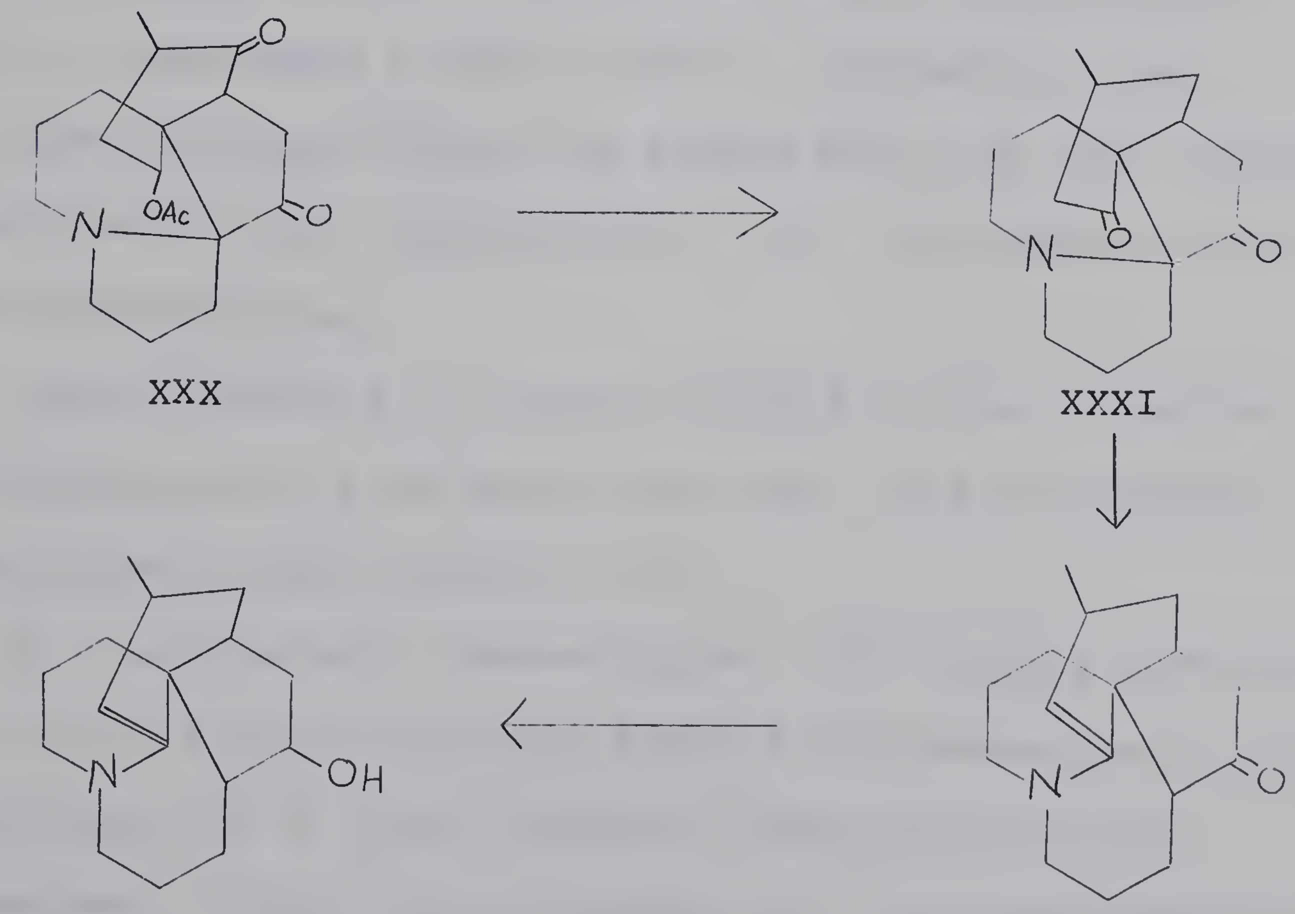
In the investigation reported in this thesis, we have made extensive use of dehydrogenation. It is therefore appropriate to make a brief reference to recent results in the application of this technique to the elucidation of structures of natural products. Valenta (39) has reviewed the general field of dehydrogenation. Two brief reviews (40, 41) deal with rearrangements that occur during dehydrogenation of terpenes and alkaloids. The usefulness of chemical or catalytic dehydrogenation lies in


OH
OH
several steps
OH
Zinc-Acetic acid
Ac
OAc
OH
Ac
OH
OH

serratinine

XXIX

CHART I



XXX

XXXI

XXXIII

XXXII

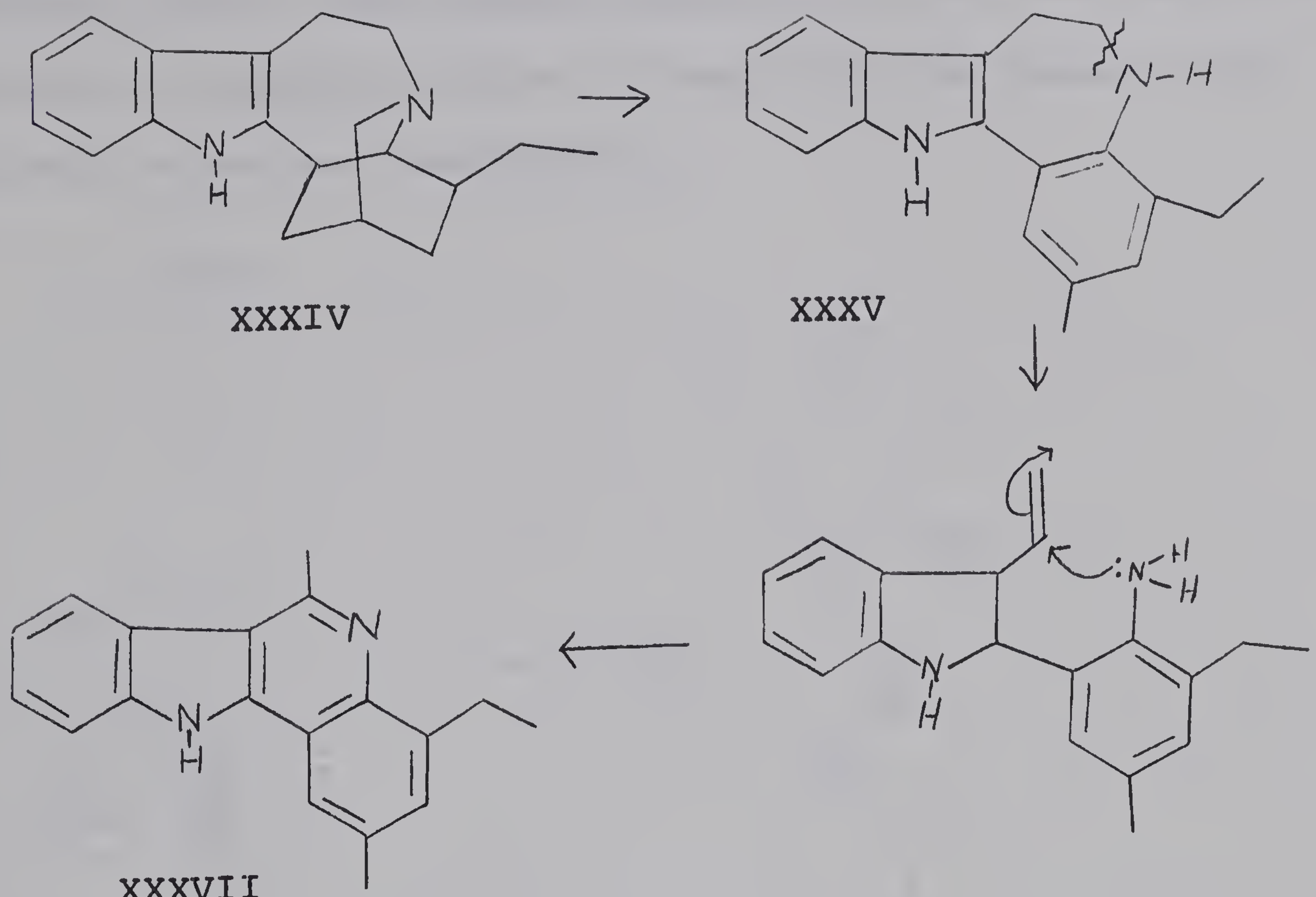
CHART II

the production of easily identifiable aromatic fragments. Classically, the often small amounts of aromatic products are identified by derivatization and by their electronic absorption spectra. The current use of gas chromatography and mass spectrometry has substantially facilitated the identification of dehydrogenation products.

The nature and substitution patterns of the dehydrogenation products gives a direct indication of the skeleton of the natural product provided that no rearrangement occurred during dehydrogenation. Rearrangements involving ring contraction, or alkyl migration do occur. For example the indole alkaloid ibogamine (XXXIV) gives the normal dehydrogenation product (XXXV), another compound XXXVI, and the rearranged product XXXVII (indoloquinoline). The chart on page 17 shows the scheme that has been suggested to rationalize the transformation (41) of ibogamine into the indoloquinoline.

Rearrangements are also involved in the formation of 7-methylquinoline from annotinine (42), and 5,7-dimethyl-1-azaanthracene from selagine (41).

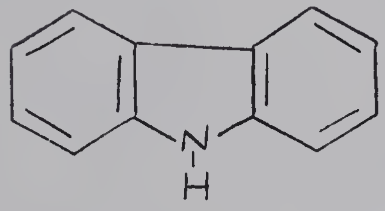
In a preliminary communication (43) Valenta and co-workers were led to assign structure XXXVIII to ormosanine by virtue of isolation of A from catalytic dehydrogenation of ormosanine. X-ray crystallography on a homo-derivative of ormosanine, jamine (44), shows that ormosanine must have the



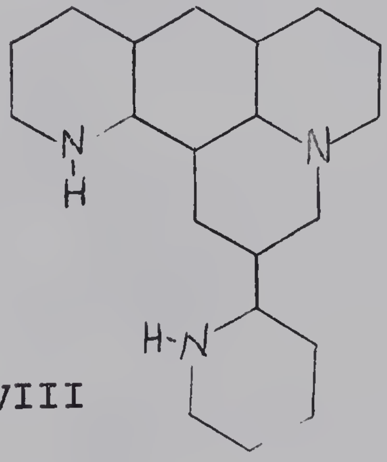
XXXIV

XXXV

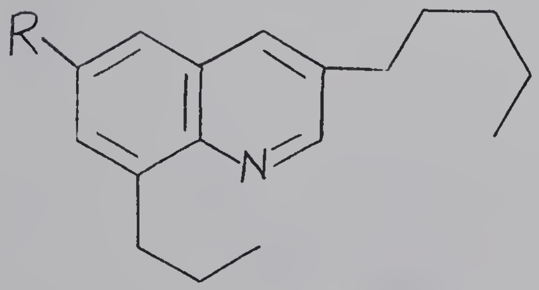
XXXVII



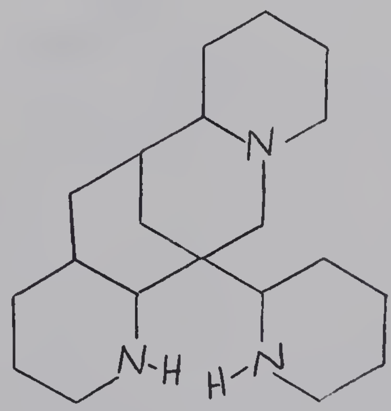
XXXVI



XXXVIII



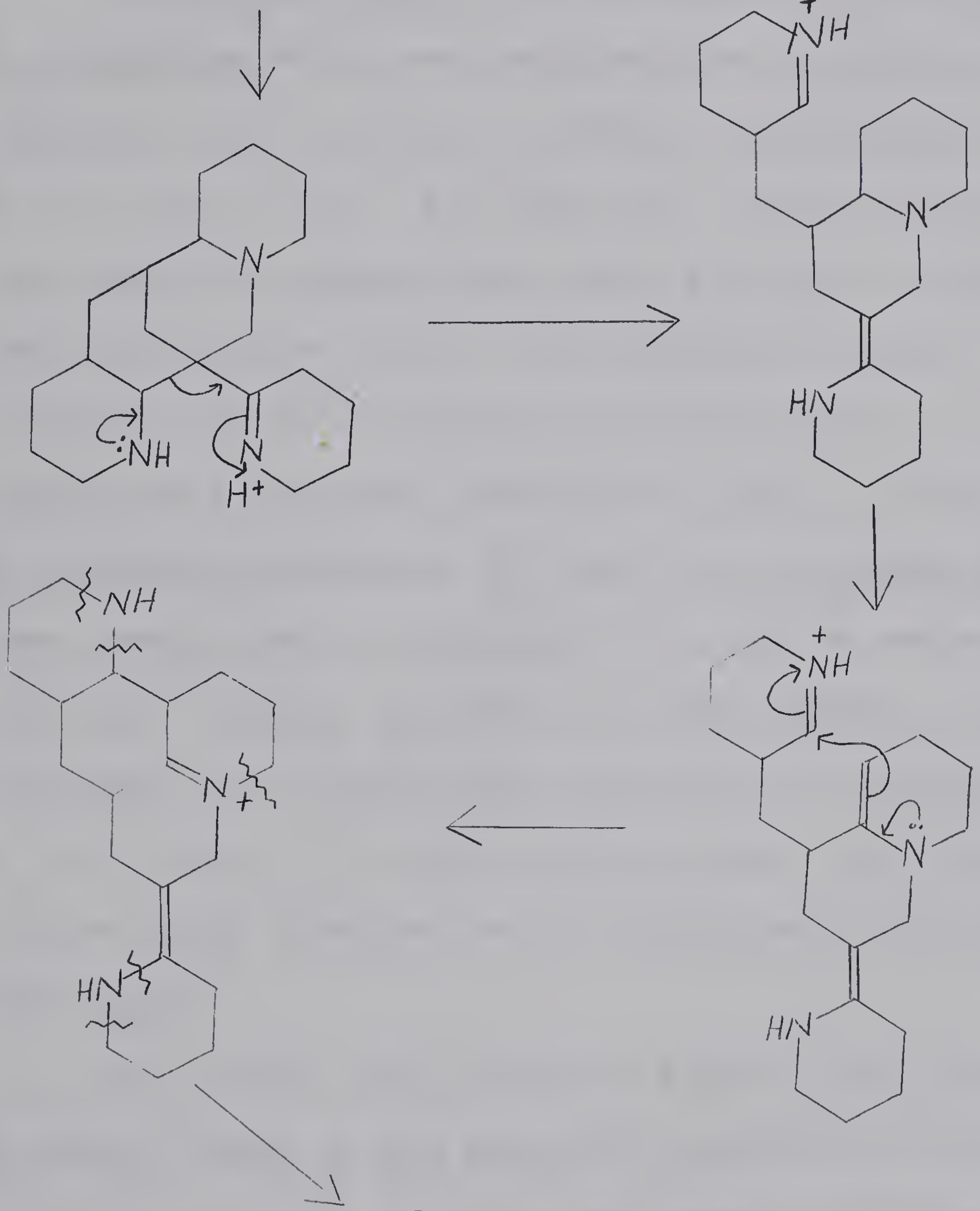
A (R = $CH_3-CH_2-CH_2-$)



XXXIX

structure XXXIX. The unusual dehydrogenative rearrangement depicted in the Chart below (45) was thus the cause of the incorrect structural assignment.

XXXIX



A

In this dissertation we report the results of our investigation of some of the alkaloids of L. lucidulum.

DISCUSSION AND RESULTS

A. The Isolation and Characterization of Weak Bases

Manske and Marion investigated the alkaloids of L. lucidulum Michx and reported the isolation of nine alkaloids (51), namely, nicotine, lycopodine, L 13, L 20, L 21, L 22, L 23, L 24, and L 25. They also reported that the amount of crystalline bases isolated constituted only one fifth of the total crude alkaloid extract. Just before we began the work reported in this thesis, it had been established in these laboratories that L 20 was 6-α-hydroxylycopodine (8) and that L. lucidulum contains some weakly basic alkaloids which had an amide band at 1620 cm^{-1} ($CHCl_3$) in their infrared spectra (46). The presence of at least seven alkaloids of unknown structure, L 13, L 21 to L 25 and the weak bases; and the availability of the plant prompted us to investigate the alkaloids of this plant.

The total crude alkaloid mixture was obtained using a method based on the procedure reported by Berezowsky (46). The dried, finely ground plant material was percolated with methanol, and the residue obtained after evaporating the methanol was then extracted successively with 6% and

10% aqueous hydrochloric acid. The aqueous solutions were extracted with ether in order to remove neutral and acidic material and basification with ammonium hydroxide followed by extraction with chloroform gave the total crude alkaloids.

The initial separation of the crude alkaloids into weak and strong bases was known to simplify the later chromatographic separation of the strong bases (46). The separation was effected by adjusting the pH of an aqueous acetic acid solution of the bases to pH8 by addition of sodium bicarbonate (47). Extraction with chloroform gave the weak bases. Basification of the aqueous solution with ammonium hydroxide led to the isolation of the strong bases. Further examination of this separation procedure revealed that lycopodine was isolated together with the weak bases, and that the weak bases could actually be extracted into chloroform from the dilute acetic acid solution. It seems that this behavior cannot be wholly due to the partial equilibration of the acetic acid between the organic and the aqueous layers since it was also found that dilute hydrochloric acid of pH 3 or 2 does not remove all the weak bases from a chloroform solution.

The desire to separate Lycopodine from the weak bases led to the exploration of other methods of separating the alkaloids into strong and weak bases. The following method has been found useful : the crude alkaloids are

dissolved in dilute hydrochloric acid and then the pH is adjusted to 5, 6 or 7 by the addition of a saturated solution of disodium hydrogen phosphate. Extraction of the pH 5-6 solution with methylene chloride gave weak bases and the addition of ammonium hydroxide to the aqueous residue and extraction gave strong bases.

Thin layer chromatography (alumina) of the weak bases indicated the presence of two major components with similar Rf values and at least six other components. Attempts to separate these components by preparative t.l.c. on alumina, column chromatography on silica-gel or florisil led to the isolation of multi-component mixtures. Continuous ether extraction of a pH 3 solution of the weak bases did not effect any separation.

However, the fact that discernible spots were formed on t.l.c. plates indicated that the components could be separated if the alumina/sample ratio was increased and t.l.c. conditions were employed for the separation. At this time, Loev and Snader (48) reported a preparative chromatographic technique with the resolvability of thin-layer chromatography. This technique, dry-column chromatography, is based on the fact that in thin layer chromatography, development is effected by movement of an eluent over dry adsorbent using only the solvent absorbed by capillary action. Thus, a column is dry-packed

and developed by allowing the solvent to move down mainly by capillary action using a solvent head of only 1-2 cm. When the solvent reaches the bottom, development is complete. The components are isolated by extrusion and slicing or further elution. Loev and Snader used alumina of activity II to III.

Since the weak bases do not give any fluorescence under ultraviolet light and the pressure exerted on extrusion might cause some band mixing, we decided to use polythene tubing columns which could be cut into 15 mm to 30 mm sections. After development of the column, careful comparative chromatographic examination of the sections led to the separation of the two major components.

We shall refer to the less polar component as lucidine-A and the more polar component as lucidine-B. Their infrared spectra are very similar and differ slightly only in the fingerprint region. They show the presence of an amide band at 1620 cm^{-1} and $N-CH_3$ at 2790 cm^{-1}. Their n.m.r. spectra have sharp signals at τ 7.84 and 7.91 and 7.95 which are assigned to $N-CH_3$ and $N-CO-CH_3$ groups Lucidine-B has two doublets in the $C-CH_3$ region in its n.m.r. spectrum whereas lucidine-A has a more complex $C-CH_3$ region. The difference in appearance of the methyl region in their n.m.r. spectra constitutes an easy method of distinguishing between lucidine-A and lucidine-B.

In this method of separation, we found that lycopodine

is isolated from two sections corresponding to Rf values of 0.8 and 0.1, and also that small amounts of lycopodine were present in some samples of lucidine-A and lucidine-B. Dry-column chromatography was thus not wholly successful and it became imperative to explore other methods of separation. We therefore turned to counter-current distribution (49). It was thought that the counter-current distribution of weak bases under suitable conditions would not only separate lycopodine from the weak bases, but might also separate the components of the weak bases.

The efficacy of counter-current distribution depends on the partition co-efficient of the compounds to be separated and on the nature of the solvents used for the two immiscible phases. The partition co-efficients of the crude weak bases between chloroform and some buffer solutions were determined and these are shown in the table on page 97

The high constancy of pH of a saturated potassium hydrogen tartarate solution over wide ranges of concentration and temperature (50) led to the use of a saturated potassium hydrogen tartarate solution as the aqueous phase.

Counter-current distribution of the weak bases between chloroform and a saturated potassium bitartarate solution over a hundred tubes with 40 ml of each phase in a Craig-Post automatic counter-current apparatus gives a distribution of bases summarized in the table following.

Fraction	Tubes	Main Components
1	0-5	1620H and luciduline
2	6-20	Luciduline
3	21-40	1620 Compounds
4	41-45	Lucidine-A, lucidine-B, 1620C and 1620D.
5	46-85	Lucidine-A, lucidine-B, 1620C and 1620D.
6	86-99	Lycopodine and two other compounds.

Fraction 4 contains more lucidine-A than lucidine-B while fraction 5 contains more lucidine-B than lucidine-A. Although counter-current distribution did not effect complete separation, repeated dry-column chromatography, of the counter-current distribution fractions led to the isolation of four new alkaloids which we have named luciduline ($C_{13}H_{21}NO$), lucidine-A ($C_{30}H_{49}N_3O$), lucidine-B ($C_{30}H_{49}N_3O$) and lycolucine ($C_{30}H_{43}N_3O$) and mixtures of alkaloids referred to as 1620 C/D and 1620 H.

B. Luciduline

Luciduline is a clear colorless oil, $[\alpha]_D$:+10°(methanol, c 0.283), which analyses for $C_{13}H_{21}NO$ (mass spectrometry). It forms a crystalline, hygroscopic perchlorate, m.p. 194-196°. These properties are very nearly the same as those reported by Manske and Marion (51) for a Lycopodium lucidulum alkaloid L 21($C_{13}H_{21}NO$), whose perchlorate has m.p. 201°. Although a direct comparison has not been possible, it seems likely that luciduline is alkaloid L 21.

The presence of absorption maxima at 2780 and 1690 cm^{-1} in the infrared spectrum ($CHCl_3$) suggest the presence of an N-methyl group and a carbonyl group respectively. Luciduline also shows absorption at 1400 cm^{-1} which is absent in luciduline-d_2(see below)and dihydroluciduline. This indicates that luciduline is a ketone and possesses a $-CH_2-\overset{O}{\overset{\|}{C}}-$ group (52). The ketonic nature of the carbonyl group is confirmed by the presence of a positive Cotton effect in the 300 mμ region in its optical rotatory dispersion curve.

The n.m.r. spectrum of luciduline (Fig.1) in deuterochloroform was very informative. The presence of the N-methyl group is confirmed by the appearance of a three proton singlet at τ 7.90. A doublet at τ 9.15 indicates the presence of a secondary methyl group in luciduline. Irradiation experiments indicate that the two low field protons at τ 6.82-7.30 are not coupled to each other and are coupled to different sets of protons.

Further information about the low field protons was

obtained by the examination of the n.m.r. spectrum of deuterated luciduline. Treatment of luciduline with deuterochloric acid acetic acid-O-d at room temperature for five days gave deuterated luciduline. Since ketones are known to readily exchange the incorporated deuterium in the inlet system of the mass spectrometer, the number of deuterium atoms exchanged was determined by examination of the mass spectrum of its sodium borohydride reduction product. Reduction of deuterated luciduline with sodium borohydride in methanol-O-d gave dideutero-dihydroluciduline as shown by the mass spectrometric shift of the molecular ion from m/e 209 to 211 and the M-17 peak from m/e 192 to 194. This clearly indicates the presence of two enolisable hydrogen atoms in luciduline and thus deuterated luciduline is luciduline-d_2.

The low field region of the n.m.r. spectrum of luciduline-d_2 in deuterochloroform (Fig.3) shows the absence of peaks which were at τ 6.96 and 7.65 in luciduline. The proton at τ 7.17, which gives rise to a doublet of doublet with splittings of 12 and 4 cps, is most likely a proton on a methylene carbon atom adjacent to the nitrogen. The proton at lowest field (τ 6.96) in the n.m.r. spectrum of luciduline must be one of the methylene protons adjacent to the carbonyl group. The large splittings associated with this peak (quartet, J=13 and 12 cps) suggest that this proton (τ 6.96) is axial whereas the other proton at τ 7.65 which has splittings of approximately 13 and 4 cps is equatorial. The probable cause of the appearance of the axial

proton at lower field than the geminal equatorial proton will be discussed later.

It is known that non-labile protons attached to carbon atoms adjacent to a basic functional group are deshielded by protonation of the basic site. A recent study by Ma and Warnhoff (53) has demonstrated that N-methyl groups can be readily detected, estimated and characterised by determining the deshielding of the N-methyl protons that occurs on changing the solvent from deuterochloroform to acidic solvents such as acetic acid-$\underline{d}_4$ (perdeuteroacetic acid) and trifluoroacetic acid.* In addition to confirming the presence of the N-methyl group it seemed that the number and nature of protons attached to the other adjacent carbon atoms could also be determined from the n.m.r. spectrum of luciduline in perdeuteroacetic acid.

The n.m.r. spectrum of luciduline in perdeuteroacetic acid is shown in Fig. 2. The N-methyl group is now τ 6.97, indicating that the nitrogen atom is basic. The region below τ 6.90 shows that apart from the N-methyl protons there are three protons on carbon atoms adjacent to nitrogen. A methine proton appears at τ 6.43 and methylene protons appear as doublets at τ 6.20 and 6.67 with a splitting of 13 cps indicative of geminal coupling. The doublet at τ 6.67 is also coupled by 4 cps to a proton at τ 7.24. It is likely that the τ 6.20 proton is equatorial and the τ 6.67 proton is axial. The above data

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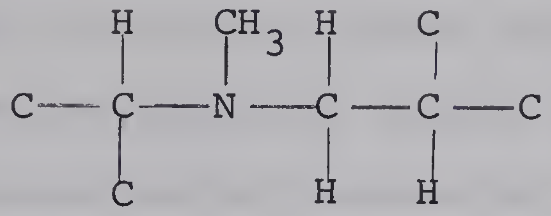
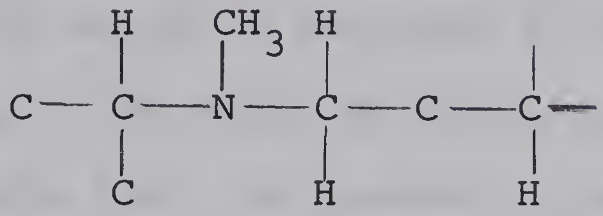
* Trifluoroacetic acid produces greater deshielding, but it was not used in this series because samples tend to decompose in this solvent.

indicate that the environment of the nitrogen is as shown in partial structure I or II



$$\mathrm{C{-}\overset{H}{\underset{C}{C}}{-}\overset{CH_3}{N}{-}\overset{H}{\underset{H}{C}}{-}\overset{C}{\underset{H}{C}}{-}C}$$

I



$$\mathrm{C{-}\overset{H}{\underset{C}{C}}{-}\overset{CH_3}{N}{-}\overset{H}{\underset{H}{C}}{-}C{-}\overset{|}{\underset{H}{C}}{-}}$$

II

The proton at τ 7.24 is probably vicinal to the methylene protons adjacent to nitrogen, and near a deshielding centre such as the carbonyl group, but it could also be on the β-carbon atom. The small splitting of 4 cps would then be explicable in terms of long range coupling through four σ bonds (54).

The relatively low frequency of the carbonyl group (1690 cm^{-1}) and the low basicity of luciduline which shows no inflexion in its titration curve when titrated with sulfuric acid in 80% methylcellosolve, seemed suggestive of the presence of an α-aminoketone grouping in the alkaloid. In order to check this possibility and also to delineate the environment of the carbonyl group, we investigated the spectral properties of dihydroluciduline, O-acetyldihydroluciduline and O-acetyldihydroluciduline-d_2.

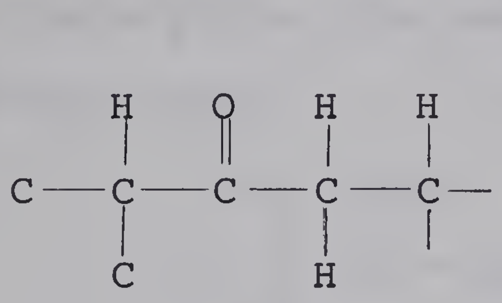
Dihydroluciduline ($C_{13}H_{23}NO$) was readily prepared by the reduction of luciduline with sodium borohydride in methanol. Dihydroluciduline, pKa 7.7 (in 80% methylcellosolve) is a stronger base than luciduline though still a relatively weak base. Its infrared spectrum in carbon tetrachloride solution showed a sharp concentration-independent absorption at 3603cm^{-1}. The n.m.r. spectrum in deuterochloroform has a broad one proton multiplet at τ 6.05 which is assigned to the proton geminal to the hydroxyl group. The width at half height and multiplicity of the peak indicate that the proton is axial and coupled to more than two protons. This proton shifts to τ 5.03 on acetylation of dihydroluciduline.

Irradiation experiments on O-acetyldihydroluciduline in deuterochloroform show that the proton at τ 5.03 is coupled to a proton at τ 7.44 by a large coupling and also to the τ 8.33 region. The n.m.r. of O-acetyldihydroluciduline in perdeuteroacetic acid is very similar to that of luciduline in perdeuteroacetic acid, except for the presence of the methine proton geminal to the acetoxy group at τ 5.00. It was demonstrated by means of decoupling experiments that none of the three protons on carbon atoms adjacent to the nitrogen are coupled to the proton at τ 5.00.

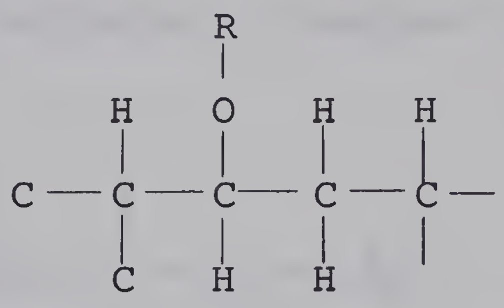
The above experiments suggest that luciduline does not possess an α-aminoketone system. This inference is also consistent with the observation that the region below τ 7.0 in the n.m.r. spectra of both luciduline and O-acetyldihydro-

luciduline in perdeuteroacetic acid are very similar to the corresponding spectra of the undeuterated compounds.

Further, the data taken together with that for luciduline indicate that O-acetylhydroluciduline has the partial structure IV($R=\overset{\mathrm{O}}{\overset{\|}{\mathrm{C}}}-\mathrm{CH}_3$) and hence luciduline itself has the partial structure III.

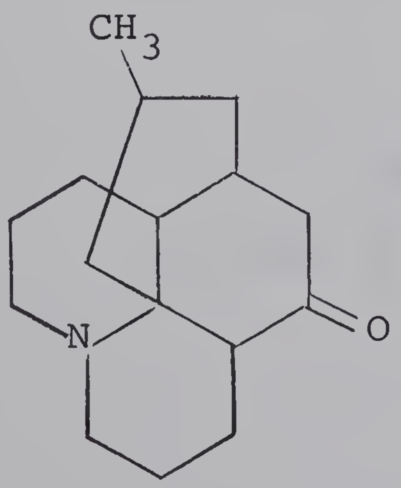


III

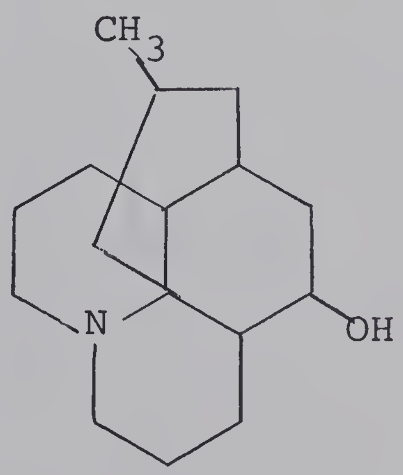


IV

Some β-aminoketones are weaker bases than the alcohols derived from them (69). For example, lycopodine 1 is a weaker base than dihydrolycopodine 2.



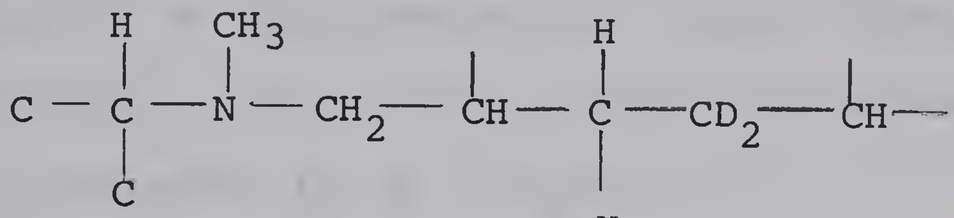
1



2

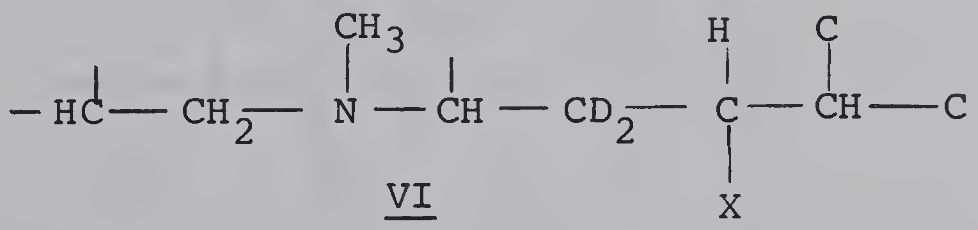
In order to test this possibility, the n.m.r. spectra of O-acetyldihydroluciduline-d_2 were examined in deuterochloroform and perdeuteroacetic acid.

If luciduline has a β-aminoketone system, then the partial structures defining the environments of the amino and ketone functions in O-acetyldihydroluciduline-d_2 can be joined together in four ways giving the partial structures denoted by V-VIII $(X=O-\overset{O}{\overset{\|}{C}}-CH_3)$ below:



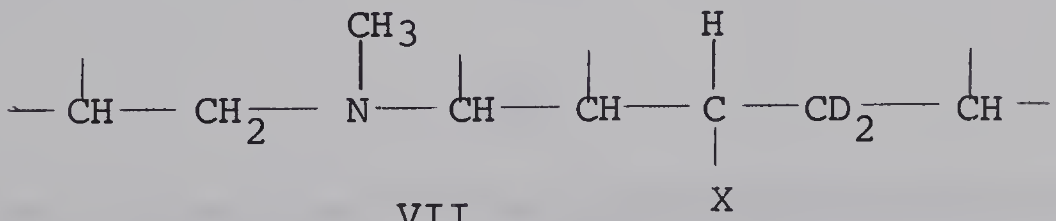
$$C-\underset{C}{\overset{H}{C}}-\overset{CH_3}{N}-CH_2-\overset{|}{CH}-\underset{X}{\overset{H}{C}}-CD_2-\overset{|}{CH}-$$

V



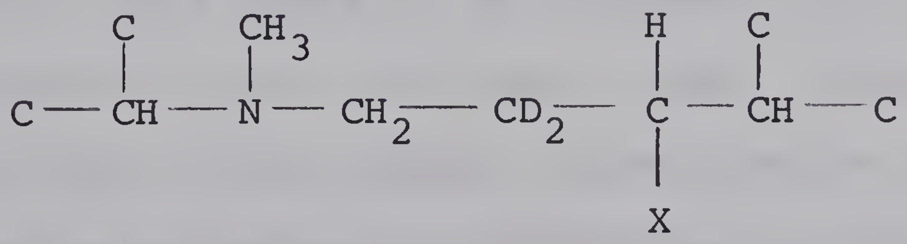
$$-H\overset{|}{C}-CH_2-\overset{CH_3}{N}-\overset{|}{CH}-CD_2-\underset{X}{\overset{H}{C}}-\overset{C}{CH}-C$$

VI



$$-\overset{|}{CH}-CH_2-\overset{CH_3}{N}-\overset{|}{CH}-\overset{|}{CH}-\underset{X}{\overset{H}{C}}-CD_2-\overset{|}{CH}-$$

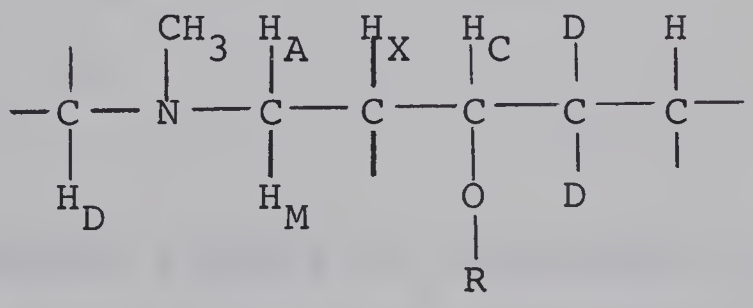
VII



$$C-\overset{C}{CH}-\overset{CH_3}{N}-CH_2-CD_2-\underset{X}{\overset{H}{C}}-\overset{C}{CH}-C$$

VIII

Since the n.m.r. spectrum of O-acetyldihydroluciduline-d_2 in perdeuteroacetic acid is very similar to the unexchanged compound structure VIII is not likely since it predicts reduced multiplicity of the adjacent methylene. If, however, O-acetyldihydroluciduline possesses the partial structure V, then the proton geminal to the acetoxy group should be split by the same proton that splits one of the methylene protons on the carbon atom adjacent to the nitrogen. Irradiation in the region of the chemical shift of this proton, H_X, should simultaneously affect the multiplicities of the protons designated H_A, H_M and H_C in partial structure IX (R = CH_3CO-)



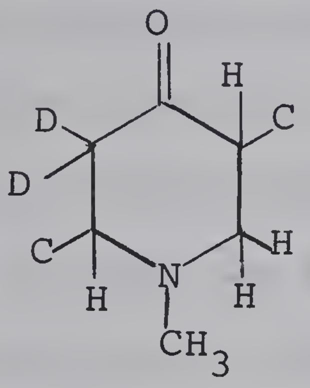
$$-\underset{H_D}{\overset{|}{C}}-\overset{CH_3}{N}-\underset{H_M}{\overset{H_A}{C}}-\overset{H_X}{C}-\underset{\underset{R}{O}}{\overset{H_C}{C}}-\underset{D}{\overset{D}{C}}-\overset{H}{C}-$$

IX

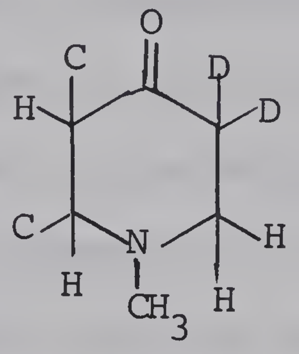
The n.m.r. spectrum of O-acetyldihydroluciduline-d_2 in perdeuteroacetic acid shows H_C as a doublet at τ 4.98 (J = 4 cps), H_M is a quartet at τ 6.81 (J=13 and 4 cps). Irradiation at τ 4.98 affects the τ 7.74 region. Simultaneous irradiation of the τ 7.74 region collapses the doublet at τ 4.98 to a singlet and removes the small splitting in the τ 6.81 peak.

These results strongly favor structure V, but may also be adduced to favor structure VI, if structure VI is a derivative of the piperidone X. Structure VII based on the N-methyl-

piperidone XI is easily excluded as a likely possibility on the same grounds as structure VIII. It must be emphasized however, that the simultaneous collapse of the τ 4.98 and 6.81 signals could be fortuitous especially as the irradiated region integrates for more than one proton.



X



XI

The molecular formula of luciduline, $C_{13}H_{21}NO$, indicates the presence of four sites of unsaturation in the molecule. One unsaturation is due to the carbonyl group and since the spectroscopic data of luciduline and dihydroluciduline do not suggest the presence of double bonds, luciduline is therefore a tricyclic alkaloid.

The mass spectrum of luciduline, Fig. 12 , is not interpretable in terms of the known mass spectral fragmentation patterns of Lycopodium alkaloids (55). We therefore decided to utilise dehydrogenation in order to gain further insight into the nature of the carbon skeleton present in luciduline. Ketones are known, on occasion, to decarbonylate on chemical or catalytic dehydrogenation and so dihydroluciduline was used in the first

dehydrogenation experiments.

A mixture of dihydroluciduline and excess selenium was heated in a sealed tube at about 300° for eight hours. The products, obtained by direct sublimation, were a mixture of two major components, A and B in the ratio of 3 to 1, and several minor components as shown by gas-liquid chromatographic analysis. The two major components have the molecular compositions $C_{12}H_{12}$ and $C_{13}H_{14}$ respectively. The ultraviolet spectrum of A indicated that it is a 2,6-disubstituted naphthalene. The fact that the molecular ion at m/e 156 is also the base peak and the presence of a relatively strong M-CH_3 peak (55% of the base peak) provide strong evidence that A is 2,6-dimethylnaphthalene. The mass spectrum and ultraviolet spectrum of the dehydrogenation product are practically identical with those of authentic 2,6-dimethylnaphthalene.

The dehydrogenation product B is also a naphthalene (UV spectrum) and is probably a trimethylnaphthalene since it has the molecular ion as the base peak, and a relatively intense peak at m/e 155 (52% of the base peak) corresponding to the loss of a methyl group.*

The absence of isolable amounts of nitrogen-containing dehydrogenation products led us to think that dehydrogenative decarbonylation of luciduline might lead to the cleavage of

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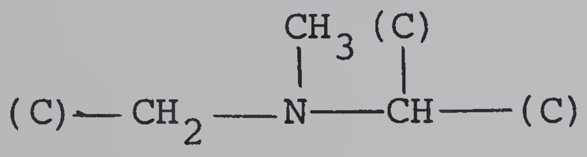
* The product B is not likely to be a propylnaphthalene or a methylethylnaphthalene since these compounds would not be expected (56) to have the molecular ion (m/e 170) as the base peak.

one of the carbocyclic rings and consequent formation of a heteroaromatic compound. We further speculated that even if decarbonylation did not occur and instead a phenolic compound were produced, elucidation of its structure would provide us with information useful in locating the position of the carbonyl group in the luciduline skeleton.

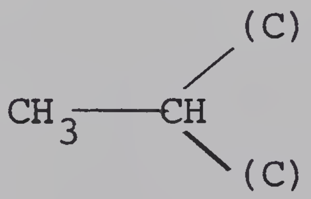
To our surprise, dehydrogenation of luciduline also gave mainly 2,6-dimethylnaphalene (u.v., i.r., and mass spectra) in remarkably high yield (above 30%). The ultraviolet spectrum of the mixture of dehydrogenation products showed practically no change on basification with sodium hydroxide or acidification with hydrochloric acid, thus indicating the absence of both phenolic and basic compounds in the mixture.

Thus far, the partial structures XII, XIII, and XIV have been definitely established.

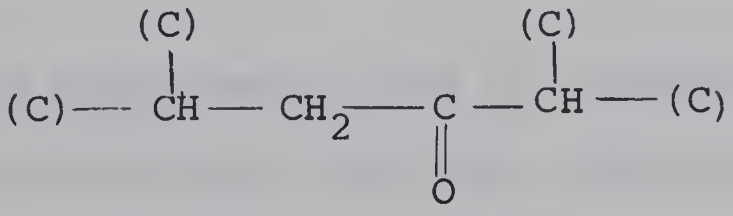


$$\mathrm{(C)-CH_2-\overset{\overset{\displaystyle CH_3}{|}}{N}-\overset{\overset{\displaystyle (C)}{|}}{CH}-(C)}$$

XII



XIII

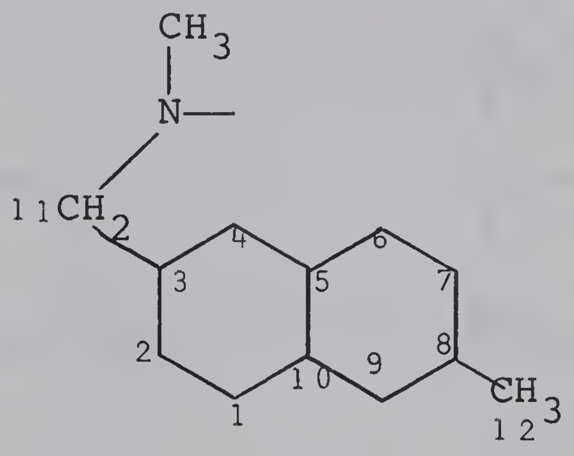


$$\mathrm{(C)-\overset{\overset{\displaystyle (C)}{|}}{CH}-CH_2-\underset{\underset{\displaystyle O}{\|}}{C}-\overset{\overset{\displaystyle (C)}{|}}{CH}-(C)}$$

XIV

The methine proton in partial structure XIV is not readily enolisable and is therefore assumed to be bonded to a bridgehead

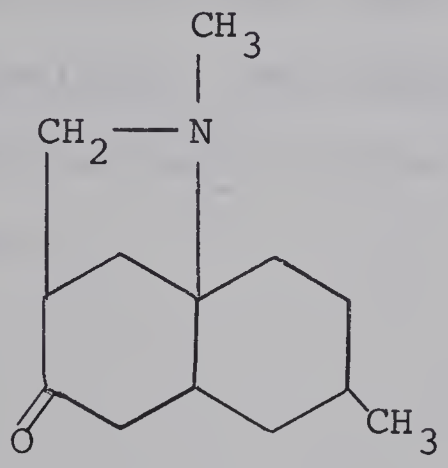
carbon atom. If it is assumed that no rearrangement occurred during dehydrogenation, then luciduline has the 2,6-dimethylnaphthalene carbon skeleton. If it is further assumed that one of the methyl groups of 2,6-dimethylnaphthalene is the same as the one originally present in luciduline and that the other methyl group arises from the cleavage of the C-N bond in $-CH_2-N-$, then the partial structure XV can be written for luciduline.



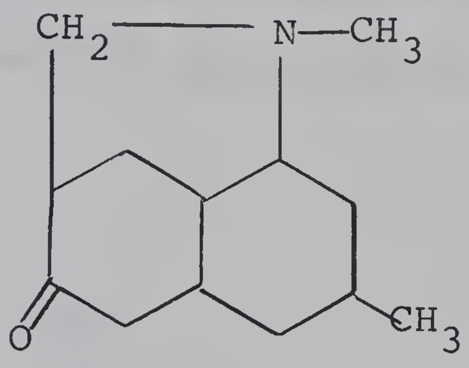
XV

Decoupling experiments done on luciduline,dihydroluciduline, O-acetyldihydroluciduline and their dideutero-derivatives indicate that the secondary methyl group is not directly attached to partial structures XII and XIV. C-7 and C-9 must be methylene carbon atoms. If the carbonyl group is placed at C-6, C-4 or C-1, it is not possible to construct a structure in which there is a a non-enolisable hydrogen on the adjacent carbon atom. On the

other hand, if the oxygen function is at C-2, then C-1 would be a methylene group and the C-3 is a bridgehead carbon atom affording the azabicyclo-(3,2,1)- octane, azabicyclo-(3,3,1)-nonane systems shown in structures XVI and XVII.



XVI



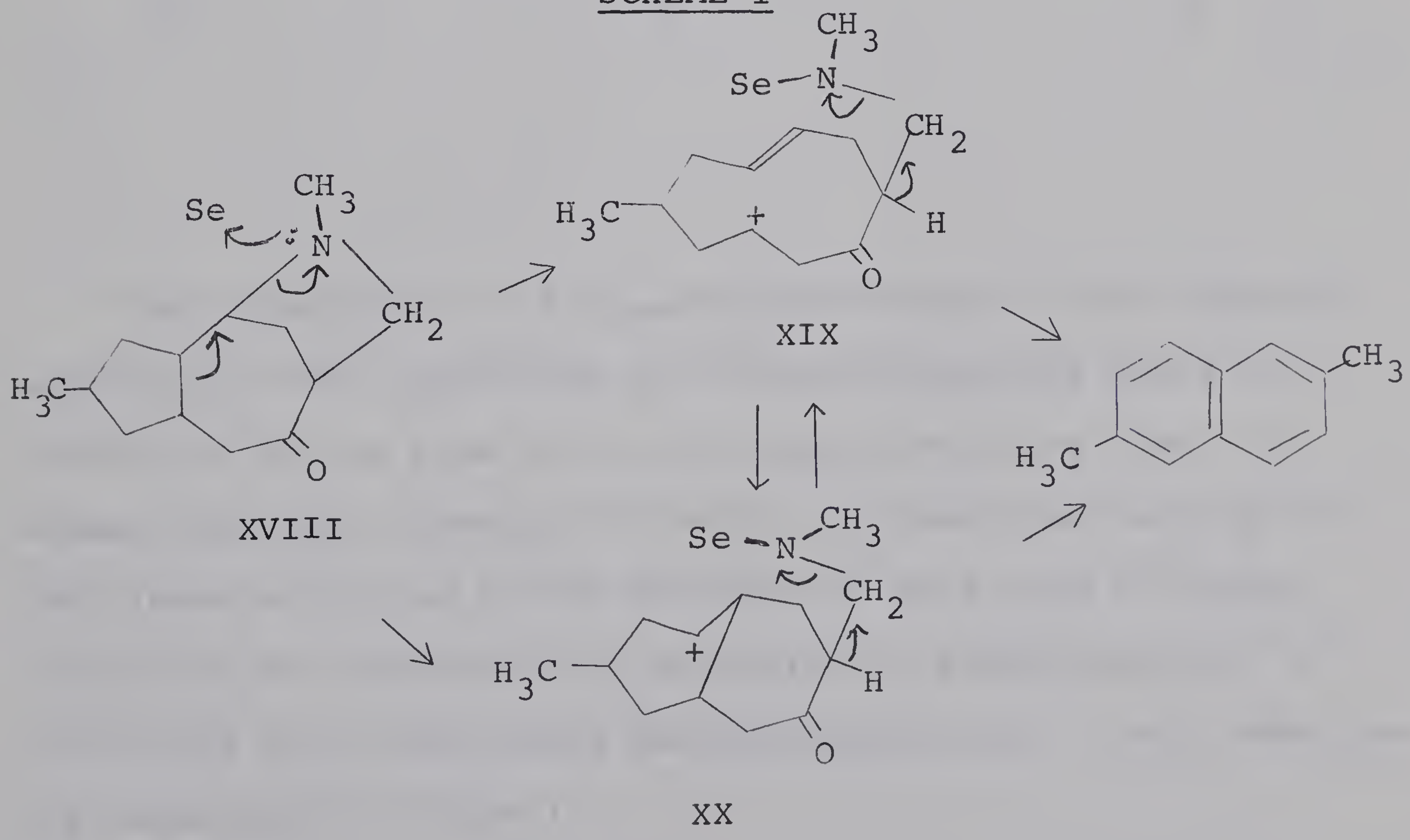
XVII

Of the two structures, only XVII incorporates the partial structure XII. Thus the structure XVII was used as a working hypothesis in further investigations of luciduline. This structure is consistent with all the results discussed above,

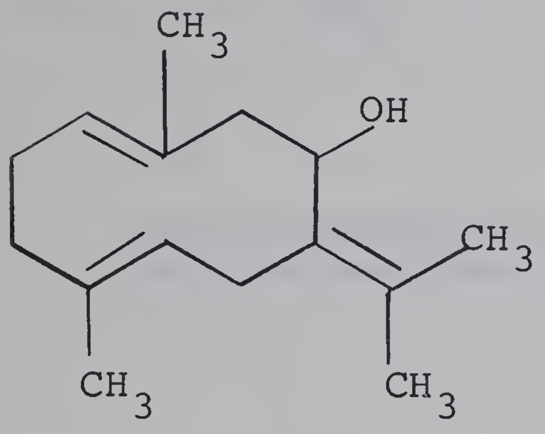
the most remarkable of which is the simultaneous decoupling of the methine proton H_C and methylene proton H_A on irradiation at H_X (Structure IX).

We have already referred to the fact that rearrangements do occur during catalytic or chemical dehydrogenation of alkaloids. It is conceivable that a naphthalene could arise from a 5,7 ring system. If 2,6-dimethylnaphthalene were obtained in this way, then one could propose structure XVIII for luciduline. The formation of 2,6-dimethylnaphthalene is then rationalized via the intermediacy of XIX or XX as shown in scheme I.

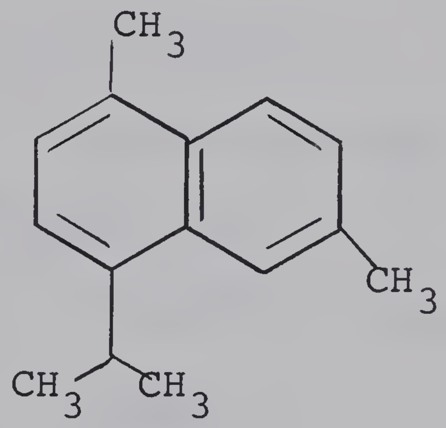
SCHEME I


Se
CH3
N
CH2
H3C
O
XVIII
CH3
Se
N
CH2
H3C
+
H
O
XIX
Se
N
CH3
CH2
H3C
+
H
O
XX
CH3
H3C

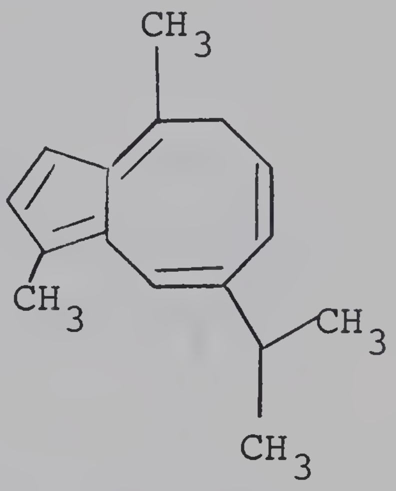
The ready cyclization of compounds with the cyclodecane skeleton to naphthalene or azulene derivatives is well known (39). For example, germacrol 3, forms the naphthalene 4 and S-guiazulene 5 on dehydrogenation (57).



3



4



5

The formation of 2,6-dimethylnaphthalene in the dehydrogenation of both luciduline and dihydroluciduline gives no indication of the size of the nitrogen-containing ring. It seemed desirable therefore to carry out reactions leading to the cleavage of ring A. The derivatives with ring A cleaved, could then be converted to a quinoline by dehydrogenation or correlated with substituted decahydroquinolines. Such reactions are summarized in chart I.

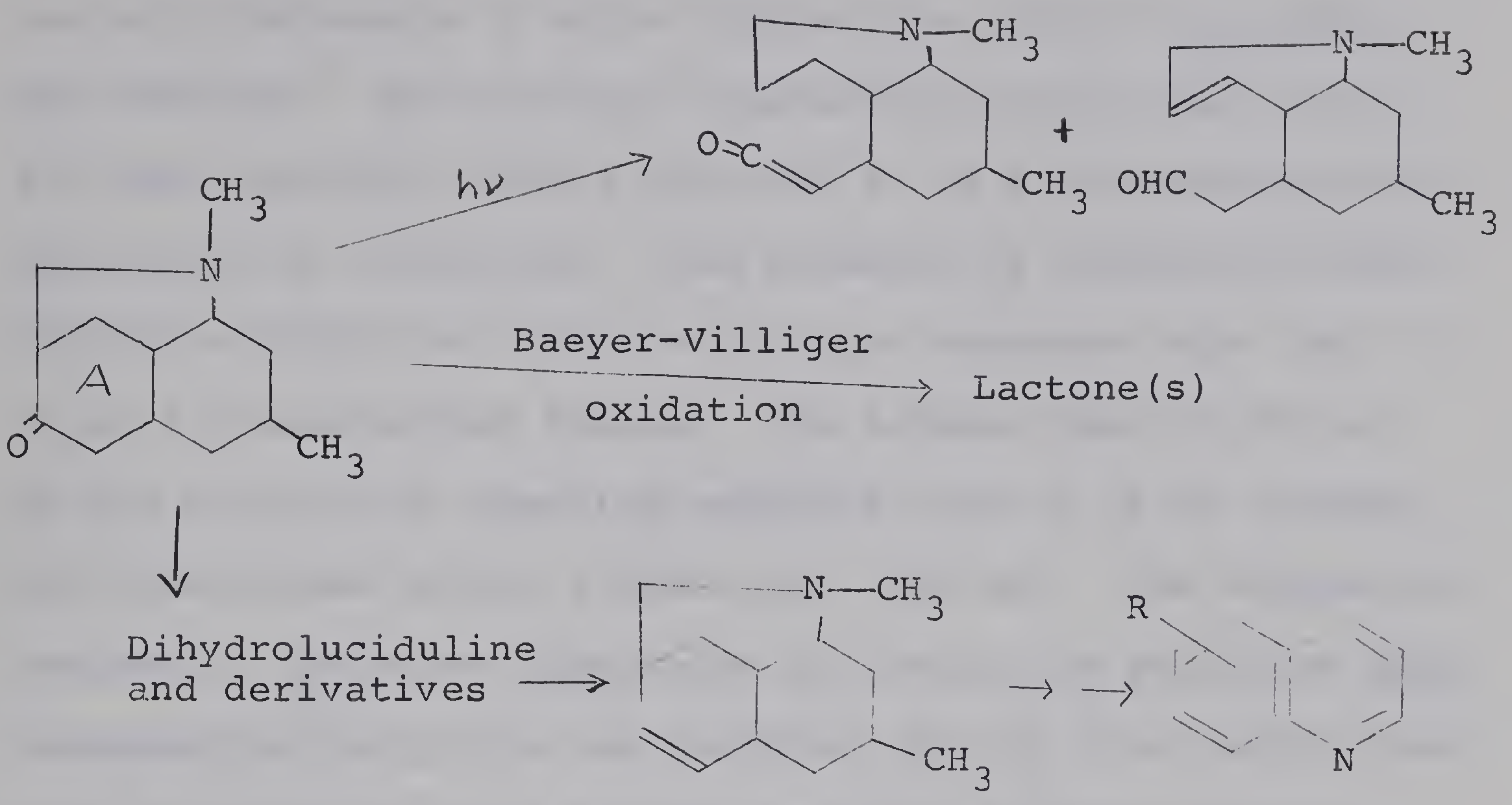

hν
$N-CH_3$
$O=C$
CH_3
+
$N-CH_3$
OHC
CH_3
CH_3
N
A
O
CH_3
Baeyer-Villiger
oxidation
Lactone(s)
Dihydroluciduline
and derivatives
$N-CH_3$
CH_3
R
N

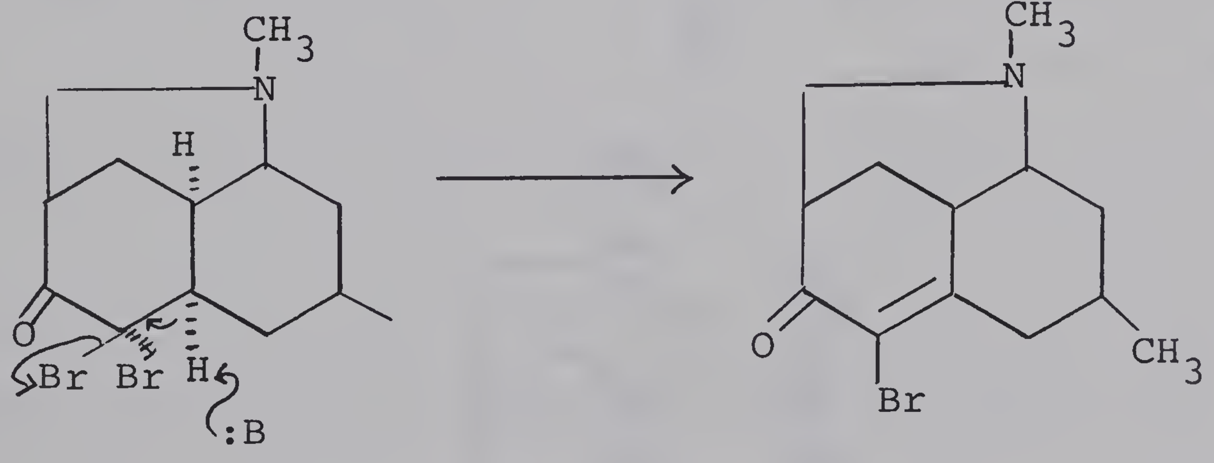
Chart I

However, when luciduline was subjected to Baeyer-Villiger oxidation or photolysis in hexane or methanol the only tractable product isolated was luciduline itself. Dihydroluciduline was resistant to dehydration with thionyl chloride in methylene chloride, benzene or pyridine, or with phosphorus oxychloride in pyridine. O-acetyldihydroluciduline was recovered unchanged on pyrolysis.

The failure of the experiments mentioned above to furnish olefinic or ring cleaved compounds led us to consider the utilization of bromination of luciduline. Dehydrobromination could lead to an αβ-unsaturated ketone which might then be cleaved by ozonolysis or periodate-permanganate oxidation. When luciduline was brominated in chloroform under acidic conditions and the product isolated by basification with aqueous

sodium bicarbonate, a white crystalline solid, $C_{13}H_{18}NOBr$, was obtained. The isotopic clusters at m/e 285 and 283 in its mass spectrum clearly indicate it is a monobromodehydro-derivative of luciduline. The presence of absorption bands at 1670 and 1603 cm^{-1} in its infrared spectrum show that it is an α,β-unsaturated ketone. The intense band at 263 mμ in its ultraviolet spectrum suggests that it is an α-bromo-α,β-unsaturated ketone (λEtOH calc. 269 mμ). The bromination product is therefore formulated as having the structure XXII. Bromodehydroluciduline was isolated (but in poor yield) even when one equivalent of bromine was used in the reaction.

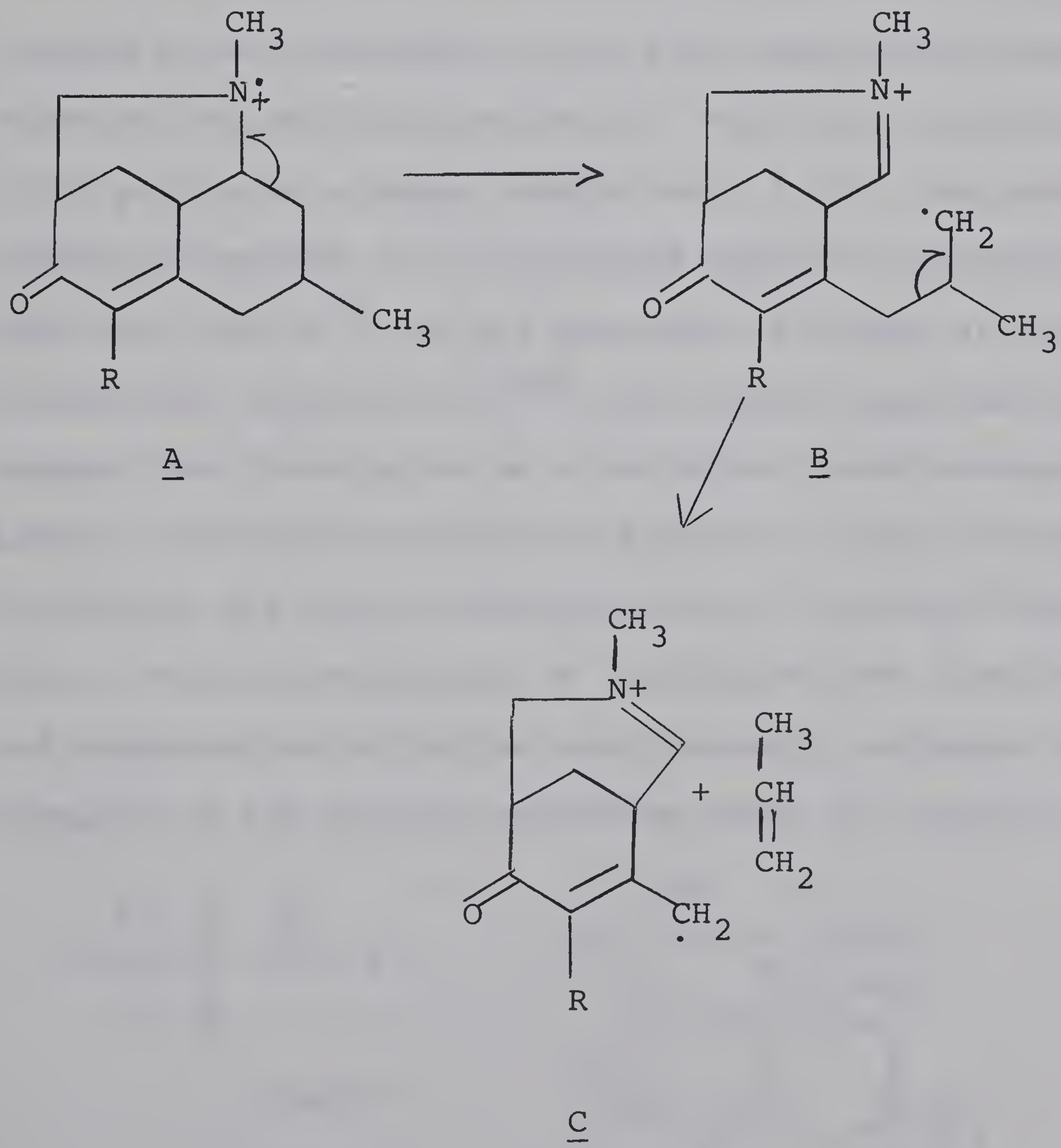
The formation of dibromoketone from a ketone when one equivalent of bromine is used, has been reported (58). It seems likely, therefore, that bromodehydroluciduline is formed by the dehydrobromination of the first-formed dibromoketone XXI.



XXI XXII

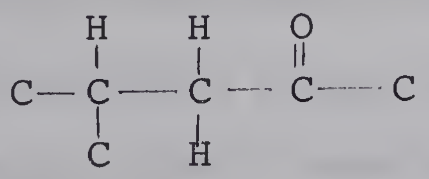
The mass spectrum of bromodehydroluciduline has prominent peaks at m/e 285 and 243. High resolution shows that the m/e 243 peak is formed from the molecular ion by the loss of C_3H_6. This fragment probably arises as indicated in the sequence A→B→C. (Scheme II, R=Br).

SCHEME II


CH3
N+
O
R
CH3
A
CH3
N+
CH2
O
R
CH3
B
CH3
N+
+
CH3
CH
CH2
O
CH2
R
C

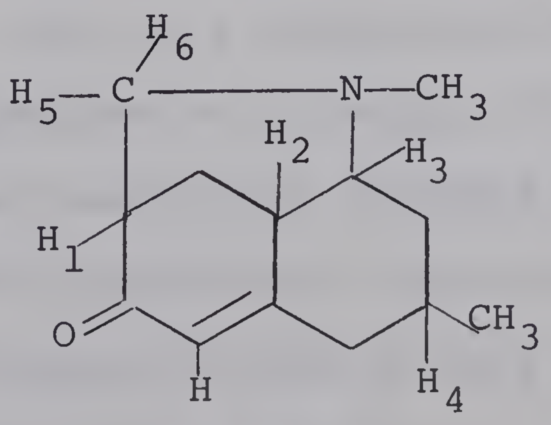
Ozonolysis and Lemieux-von Rudloff oxidation (59) of bromodehydroluciduline led to the formation of intractable mixtures. This route to ring-cleaved derivatives was abandoned and we decided to oxidize luciduline with selenium dioxide.

Work carried out in these laboratories (7) has shown that lycopodine is converted, albeit in low yields, to a diosphenol on oxidation with selenium dioxide. However, luciduline gave no diosphenol on selenium dioxide oxidation. Instead it was oxidized to the α,β- unsaturated ketone XXIV as shown by the following evidence. The n.m.r. spectrum has a vinyl proton as a sharp singlet at τ 3.98. The presence of absorption maxima in its infrared spectum (chloroform) at 1668 and 1628 cm^{-1} and the presence of a band at 242mμ in its ultraviolet absorption(λ^{EtOH} calc.244mμ) spectrum amply support its formulation as a β-disubstituted αβ-unsaturated ketone. Its mass spectrum is similar to that of bromodehydroluciduline and can be rationalized in a similar fashion (Scheme II R=H). The transformation of luciduline into dehydroluciduline and bromodehydroluciduline unequivocally indicates the presence of the partial structure XXIII in luciduline.



$$C-\underset{C}{\overset{H}{C}}-\underset{H}{\overset{H}{C}}-\overset{O}{\overset{\|}{C}}-C$$

XXIII



XXIV

The n.m.r. spectrum of dehydroluciduline (Fig. 4) is noteworthy in that the two sets of similar one proton signals centred at $\tau 7.12$ and 8.31 are coupled to the same region of the spectrum, but are not coupled to each other.

Thus, irradiation at $\tau 7.62$ collapses both the $\tau 7.12$ and 8.30 multiplets to simple doublets and strong irradiation at $\tau 7.83$ collapses the $\tau 8.30$ signal to a sharp singlet and the $\tau 7.12$ signal to a singlet with a shoulder. These results are difficult to rationalize in terms of structure XXIV, but it is possible that the first order analysis which leads one to infer that each of the signals constitutes a doublet, (J=11 cps) further split into a triplet (J=2.5 cps) is misleading. The observed multiplicity probably arises from a combination of both vicinal and long range coupling. In terms of structure XXIV, the proton at $\tau 7.12$ could be any one of the protons H_1,H_3,H_5 or H_6. Decoupling experiments demonstrate that the proton at $\tau 8.30$ is not H_4, but it could be the allylic proton H_2. It seems possible, however, that H_1 and H_2 could be the protons with signals at $\tau 7.12$ and 8.30.

Selenium dioxide oxidation is sometimes accompanied by skeletal rearrangements as illustrated by the oxidation of iso-α-amyrenonyl acetate (60) to a product in which one of the methyl groups has undergone a 1,2 shift. It seemed possible that a rearrangement could have occurred in the selenium dioxide oxidation of luciduline. Small scale catalytic hydrogenation of dehydroluciduline gave products whose mass spectrum is the same as

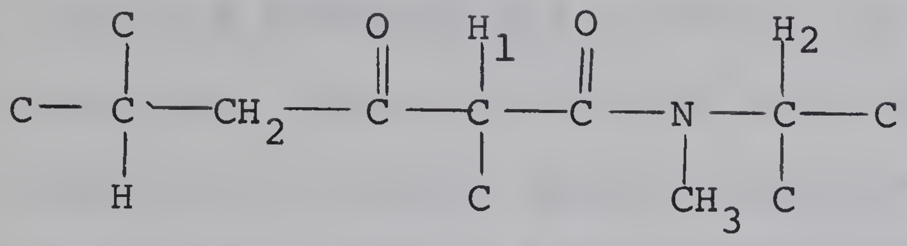
that of dihydroluciduline. It is therefore assumed that no rearrangement occurred during the oxidation of luciduline to dehydroluciduline.

The difficulties encountered in the rationalization of the n.m.r. spectrum of dehydroluciduline and to some extent that of bromodehydroluciduline, prompted us to undertake experiments to establish the way in which the partial structures XII and XIV (page 35) are joined and to ascertain whether the nitrogen is in a five-, six-, or large-membered ring.

When luciduline was oxidized with potassium permanganate in acetone, luciduline lactam, $C_{13}H_{19}NO_2$ was formed in low yield. Its infrared spectrum in chloroform has absorption maxima at 1730 and 1640 cm^{-1} which are assigned to the ketone and lactam groups respectively. The absorption of the ketone at a higher frequency than in luciduline is, by analogy to other β-dicarbonyl systems (61), ascribed to its being in a 1,3-relationship with another carbonyl group. The absorption of the lactam carbonyl group at 1640 cm^{-1} indicates that the nitrogen is in a six- or large-membered ring and thus luciduline does not have the structure XVIII.

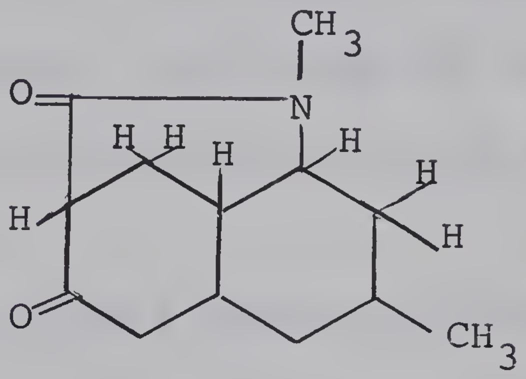
The n.m.r. spectrum of luciduline lactam shows three low field signals, namely, a one-proton quartet at τ 6.36, a one-proton multiplet at τ 6.72 and a three-proton singlet at τ 7.04 (N-methyl). The peaks at τ 6.36 and 6.72 are assigned to the methine protons designated H_1 and H_2 in the

partial structure XXV.



$$\mathrm{C{-}\overset{C}{\underset{H}{C}}{-}CH_2{-}\overset{O}{\overset{\|}{C}}{-}\overset{H_1}{\underset{C}{C}}{-}\overset{O}{\overset{\|}{C}}{-}\underset{CH_3}{N}{-}\overset{H_2}{\underset{C}{C}}{-}C}$$

XXV

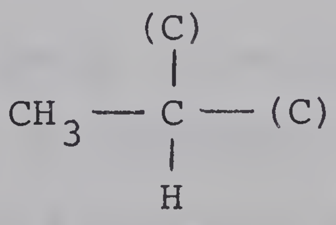


XXVI

Since luciduline lactam is presumed to have the structure XXVI, the multiplicities of these protons are readily explained by the structure proposed for luciduline lactam. H_1 is split into a quartet by the adjacent non-equivalent methylene protons. The more complex multiplet assigned to H_2 is caused by the vicinal coupling to three adjacent protons.

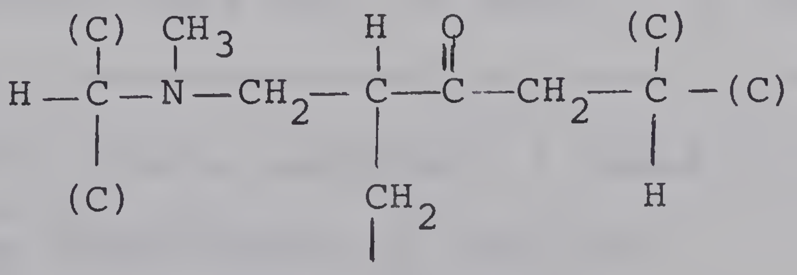
Oxidation of luciduline lactam with selenium dioxide affords dehydroluciduline lactam $C_{13}H_{17}NO_2$. Dehydroluciduline lactam is an α,β-unsaturated ketolactam as shown by absorption at 1685 and 1640 cm^{-1} in its infrared spectrum and at 230 mμ in its ultraviolet spectrum. Dehydroluciduline has a band at 242 mμ, the blue shift to 230 mμ in dehydroluciduline lactam is most likely due to the effect of the lactam carbonyl beta to the chromophore. The n.m.r. spectrum of the dehydroketolactam shows a vinyl proton at τ4.16 and the deshielded protons at τ 6.10 and 6.76 correspond to H_1 and H_2 in structure XXV.

The results discussed above establish the partial structures XIII and XXVII. These partial structures define the environment of ten of the thirteen carbon atoms in luciduline. These results are completely consistent with the structure XVII proposed for luciduline.



$$CH_3-\overset{\text{(C)}}{\underset{\text{H}}{C}}-\text{(C)}$$

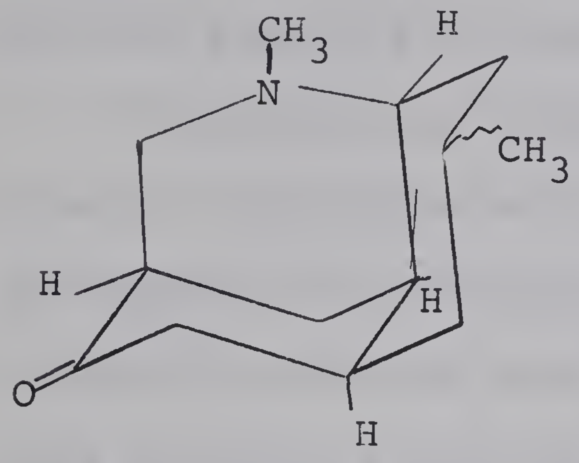
XIII



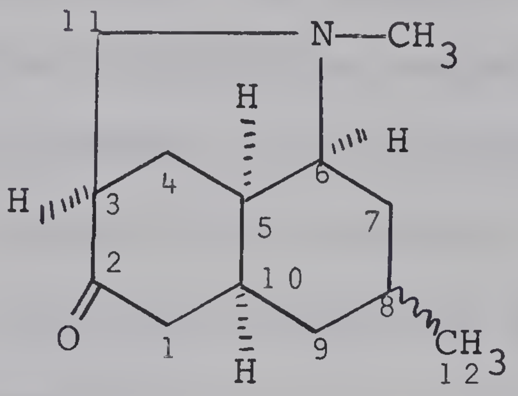
$$H-\underset{\text{(C)}}{\overset{\text{(C)}}{C}}-\overset{CH_3}{N}-CH_2-\underset{CH_2}{\overset{H}{C}}-\overset{O}{\overset{\|}{C}}-CH_2-\underset{H}{\overset{\text{(C)}}{C}}-\text{(C)}$$

XXVII

The structure derived for luciduline requires that both ring fusions be cis, and this defines the relative stereochemistry at all asymmetric centres except C-8.



XXIX



XXVIII

Thus luciduline has the relative stereochemistry shown in structure XXXVIII*. If it is assumed that all rings have chair conformations, then luciduline can be represented by structure XXIX.

Since the methyl group is conformationally larger than the nitrogen lone pair of electrons, the methyl group attached to nitrogen is assumed to be in the equatorial position whereas the lone pair is axial. In the all-chair

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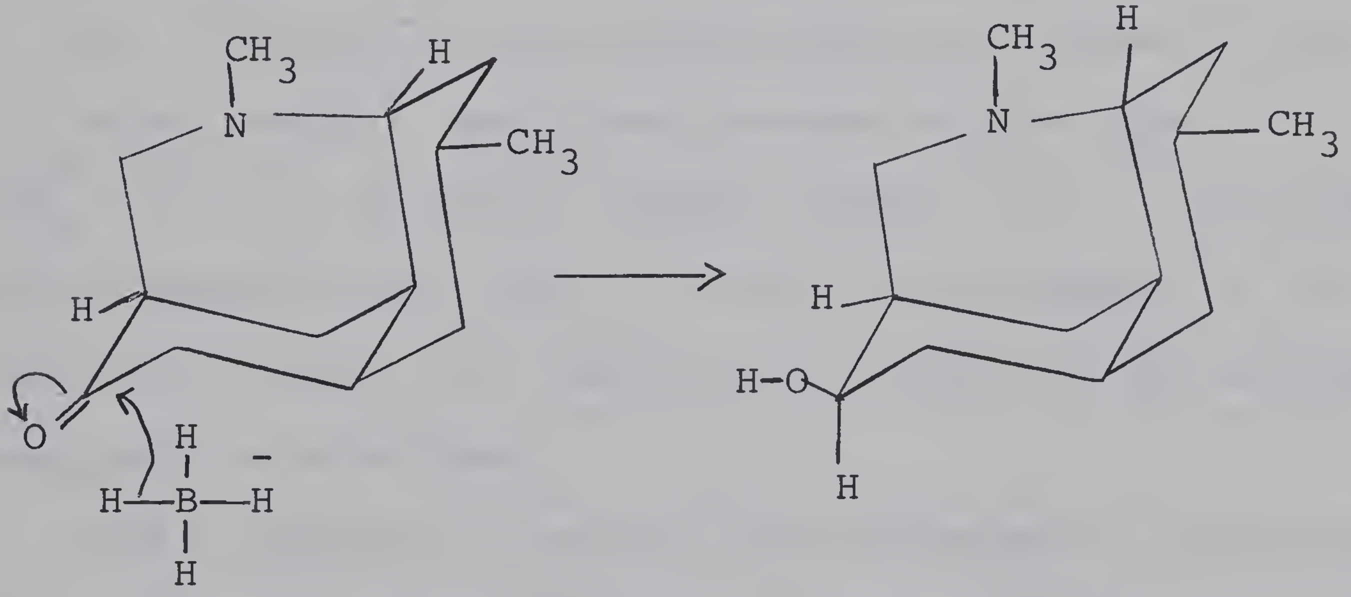
* The numbering system employed is based on the scheme used in the nomenclature of decalins.

conformation this orientation of the N-methyl group requires that the lone pair point towards the concave and more hindered side of the molecule and this would help to explain the low basicity of luciduline.

The exclusive formation of only one compound on reduction of luciduline with sodium borohydride suggests that one side of the molecule is hindered. The proposed structure correctly predicts that the reagent will approach from the less hindered side with the resultant formation of an equatorial alcohol (see scheme III).

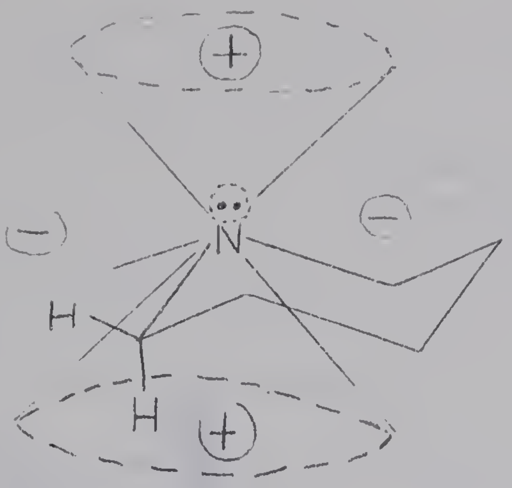


Scheme III

The orientation of the nitrogen lone pair seems to affect the chemical shifts of the C-1 axial protons in luciduline and O-acetyldihydroluciduline. As already mentioned, careful comparison of the n.m.r. spectra of

luciduline and luciduline-d_2 shows that deuterium exchange removes protons at τ 6.96 and 7.65. The proton at τ 6.96 is assigned the axial orientation because it has two large splittings whereas the one at τ 7.65 has one large and one small splitting. In the absence of other effects, methylene protons adjacent to ketogroups occur in the τ 7.4-7.8 region and the equatorial proton usually, but not invariably (62), resonates at higher field than the axial proton. In O-acetyldihydroluciduline one of the protons which is exchanged has large splittings and appears at τ 7.44. This peak has therefore been attributed to the C-1 axial proton. Methylene protons of the type $-CH_2 - C - O - R$ usually appear around τ 8.1. It is therefore suggested that the C-1 axial proton appears at low field as a result of a deshielding effect of the nitrogen lone pair of electrons.

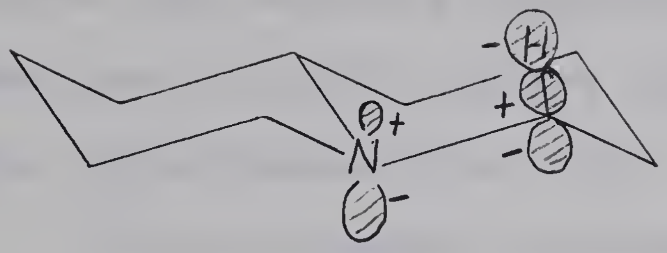
This suggestion implies that the magnetic anisotropy of the tertiary amine nitrogen lone pair is paramagnetic, and yet it has been suggested (63, 64) that the nitrogen lone pair exerts a diamagnetic effect similar to acetylene as indicated in Structure 6.



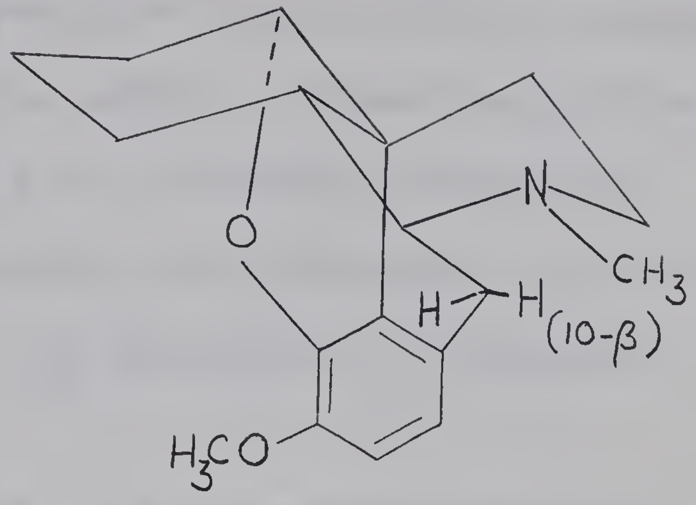
6

To our knowledge, this suggestion that the nitrogen lone pair has a diamagnetic anistropic effect is based solely on the observation that axial protons on carbon atoms adjacent to nitrogen appear at higher field than equatorial protons in quinolizidine and other compounds with similar stereochemistry. It has been suggested (64), however, that in quinolizidines the shielding of axial protons on α-carbon atoms is explained by the participation of the lone pair in a σ^{*} C-H_{ax} orbital on the adjacent carbon as shown in structure 7. Thus the shielding of axial protons in quinolizidines is explained in terms of a charge transfer effect.

A study of the n.m.r. spectra of some morphine alkaloids indicates that tertiary amino nitrogen has a paramagnetic anistropic effect on proton resonance (65). For example the C-10-β proton in 8 is shifted by 0.35 p.p.m. downfield compared to the methylene protons of



7



8

ethylbenzene largely due to the magnetic anisotropy of the nitrogen.

It is thought likely (65), that similar deshielding effects explain the observation (66, 67) that axial methyl groups have resonances at lower field than equatorial methyl groups in methylquinolizidines. The n.m.r. results obtained in the study of the luciduline series are also readily rationalized if the nitrogen lone pair exerts a paramagnetic anisotropic effect.

Luciduline displays a positive Cotton effect in its optical rotatory dispersion curve with a peak at 308 mμ and a trough at 268 mμ. The octant diagrams for the two antipodes of luciduline (XXIXA and XXIXB) are shown on page 54.

When the Octant Rule (68) is applied to these two structures, XXIXA predicts a positive Cotton effect whereas XXIXB predicts a negative Cotton effect. Since luciduline has a positive Cotton effect in its rotatory dispersion curve, the absolute stereochemistry of luciduline is that depicted in XXIXA, except for the uncertainty concerning the stereochemistry at C-8.

In the diagrams on page 54, it is assumed that all rings are in the chair form and that the secondary methyl group is equatorial. It is still possible, however, that the secondary methyl group might be axial even in the all-chair conformation. It must also be emphasized, however, that the above evidence does not exclude the possibility of boat conformations for one or more rings in Structure XVII.

In order to establish the absolute stereochemistry at C-8 and resolve some of the problems arising from spectral data, we have prepared the p- bromobenzoyl derivative of dihydroluciduline for X-ray analysis*.

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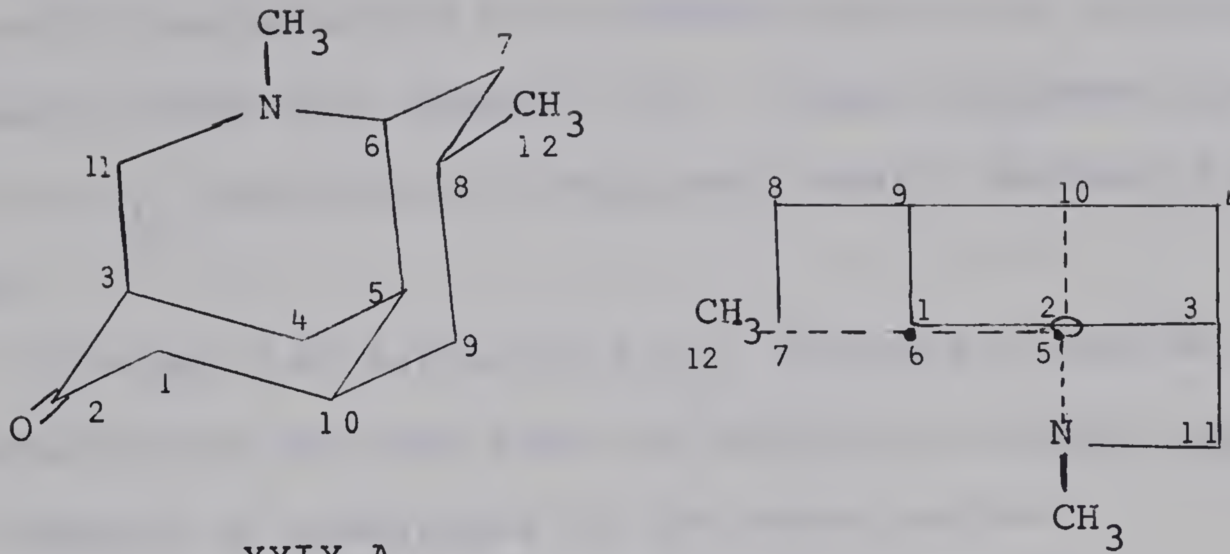
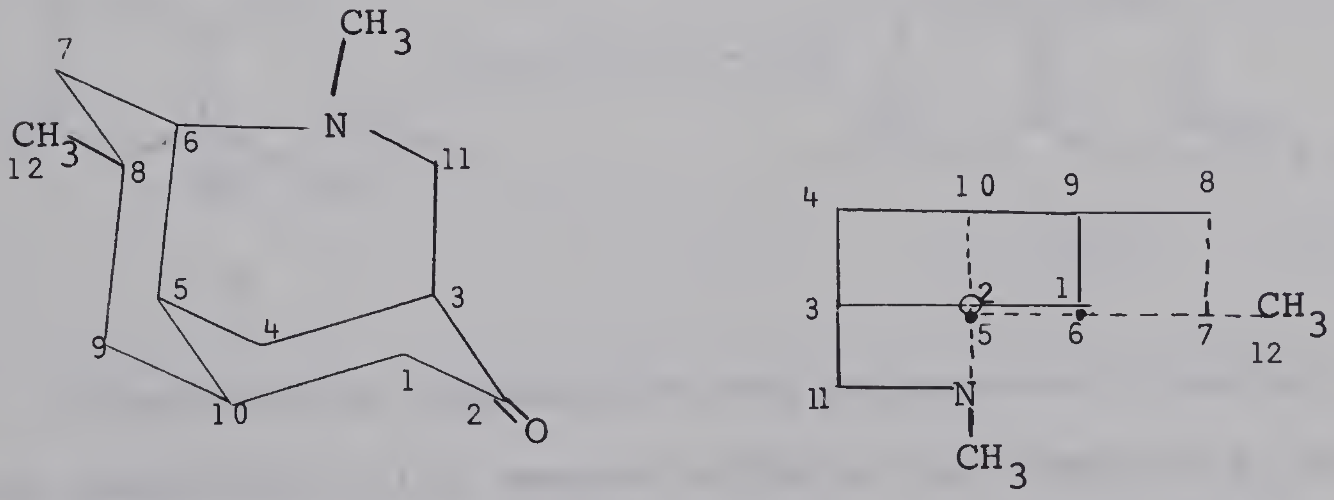
* The X-ray analysis is being done by Dr. Norio Masaki in these laboratories.


CH3
N
O
CH3
12
1
2
3
4
5
6
7
8
9
10
11

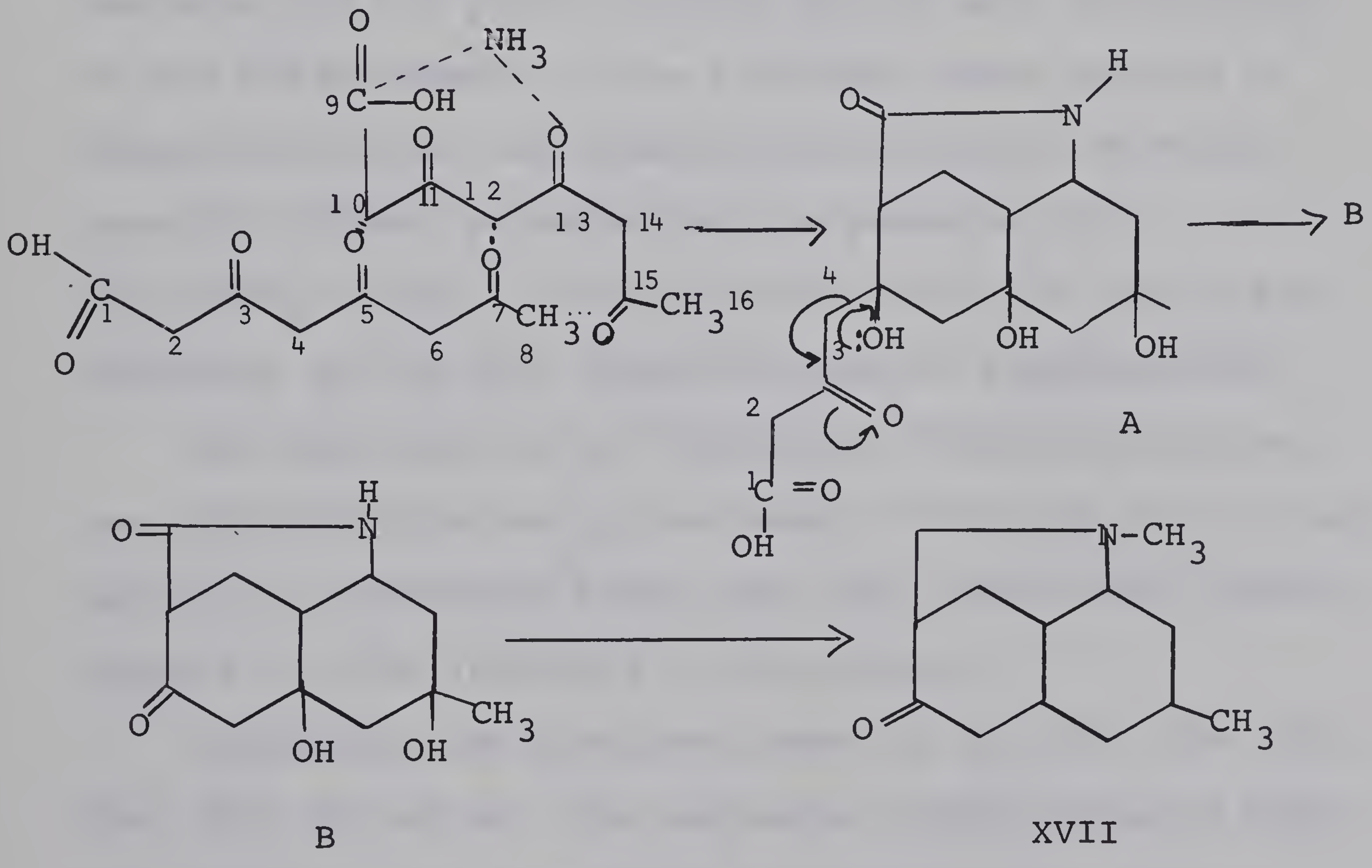
XXIX A


CH3
N
O
CH3
12
1
2
3
4
5
6
7
8
9
10
11

XXIX B

In spite of the lack of direct experimental evidence, the biogenesis of all known Lycopodium alkaloids is satisfactorily explained by the condensation of two polyketo-octanoic acids with ammonia (33). These condensations lead to C_{16} compounds or compounds readily derived from them.

Although luciduline is a C_{13} alkaloid it can be hypothetically derived from two polyketo-octanoic acids and ammonia as summarized in the chart below:



Condensation between the two polyketo-acid chains and combination with ammonia affords the compound A, the retroaldolisation of which gives the C_{12} compound B.

Luciduline may be formed from B by suitable adjustment of of oxidation levels and methylation of the nitrogen.

Mass spectrometry has been extensively used in the elucidation of the structures of natural products (70). In the course of the elucidation of the structure of luciduline, mass spectral data were extensively used to detect or confirm the presence of functional groups and to determine the molecular composition of various derivatives of luciduline. Although the novelty of the skeleton did not permit greater use of mass spectrometry in the establishment of the structure, mass spectra of dehydroluciduline and bromodehydroluciduline certainly provide indirect evidence for the presence of a CH_3-CH-CH_2- group in luciduline by virtue of the loss of propylene as the most important mode of fragmentation.

The mass spectra of luciduline, dihydroluciduline, and dihydroluciduline-d_2 are shown in Figures 12 to 14 and partially interpreted since such data constitutes further support for the structure of luciduline.

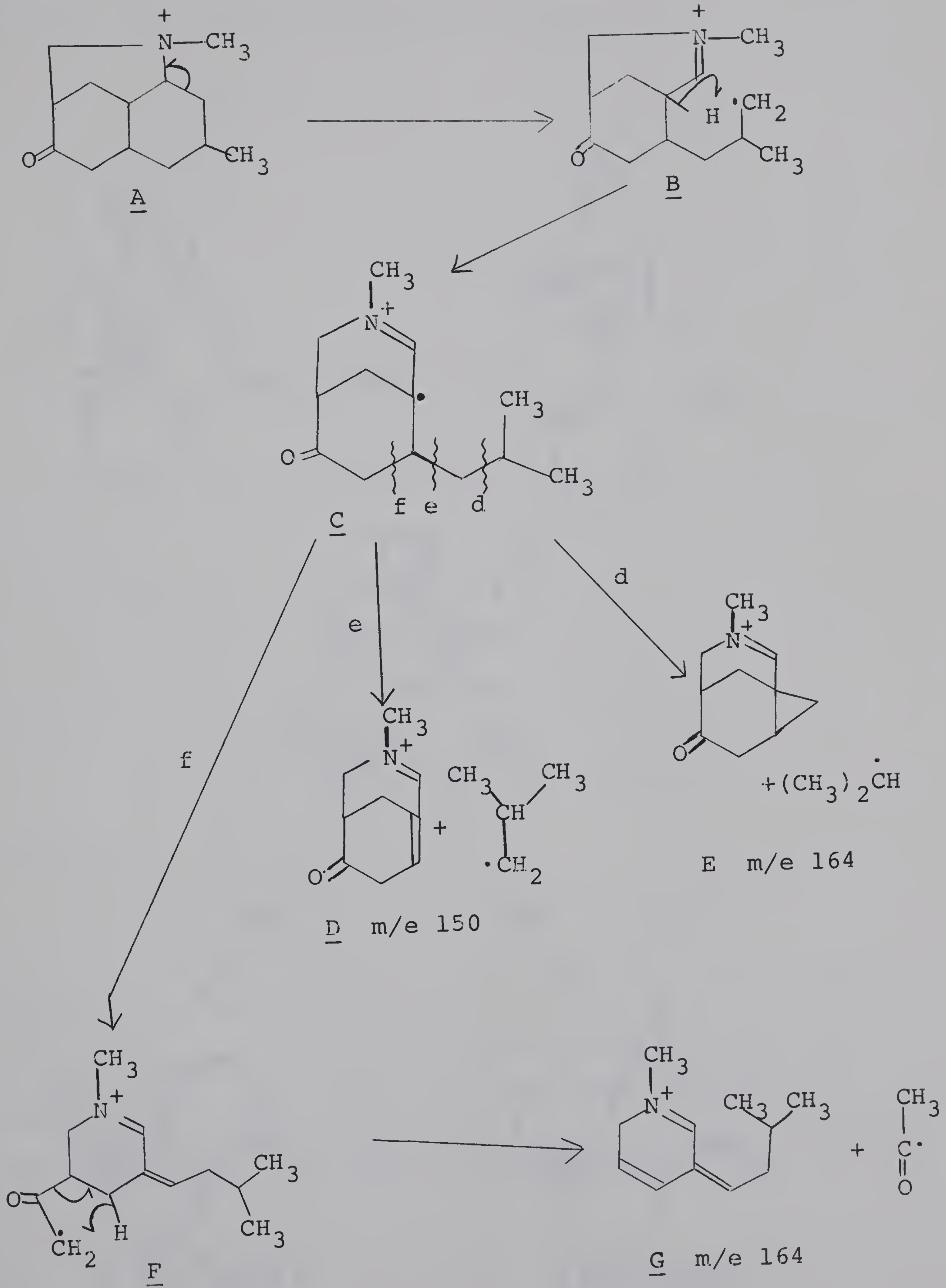
Luciduline has prominent peaks at m/e 207, 206, 192, 164, 150, 96 and 44. The molecular compositions of some of these ions are summarized in the following table.

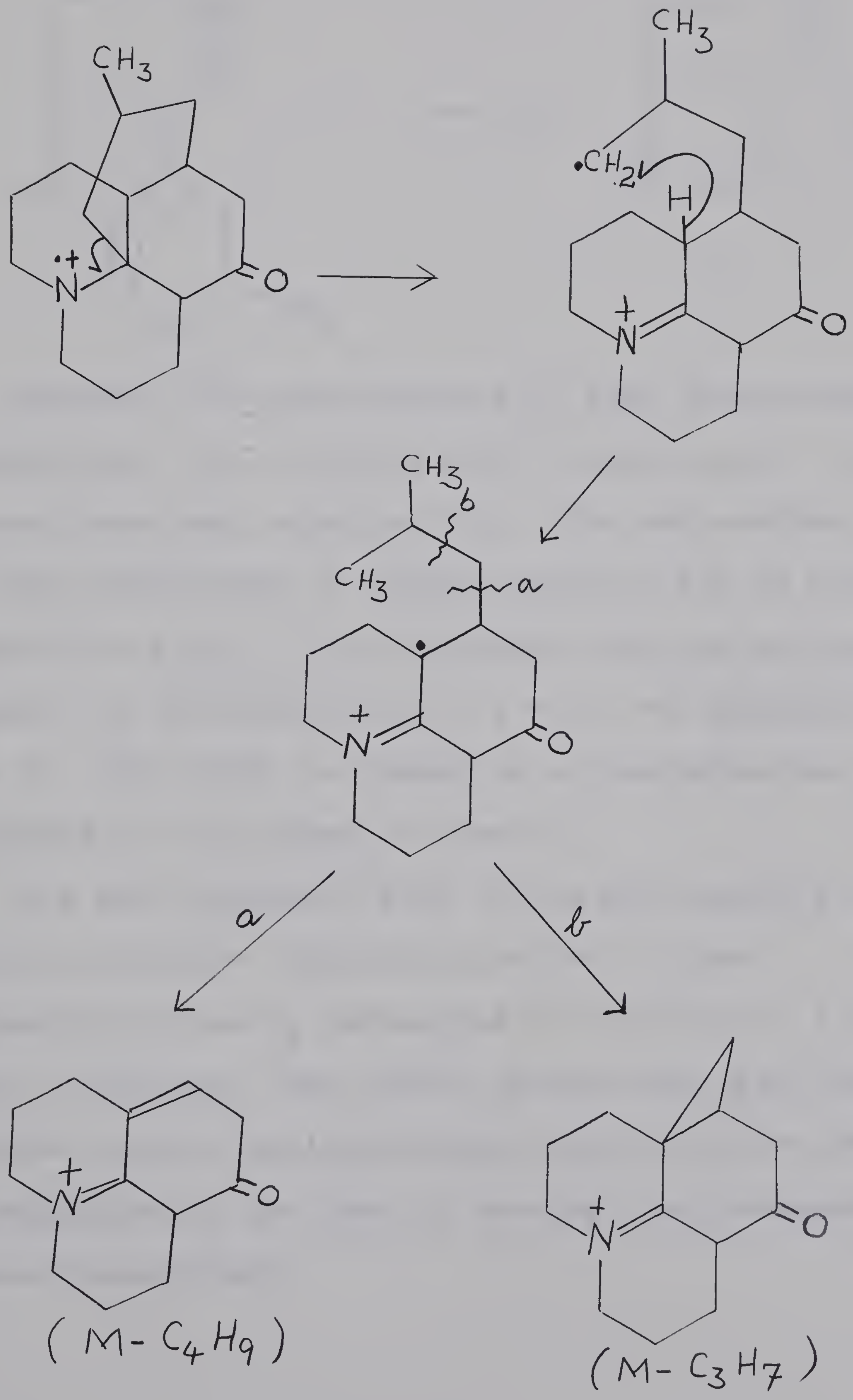
Molecular Composition	Mass Calculated	Mass Observed
$C_{13}H_{21}NO$	207.1623	207.1630
$C_{11}H_{18}N$	164.1439	164.1439
$C_{10}H_{14}NO$	164.1075	164.1076
$C_{10}H_{16}N$	150.1282	150.1281
$C_9H_{12}NO$	150.0919	150.0919
$C_6H_{10}N$	96.0813	96.0810
C_2H_6N	44.0504	44.0500

The presence of doublets at m/e 164 and 150 suggests the existence of at least two pathways to the m/e 164 and 150 ions. The formation of some of the prominent fragments is rationalized by the fragmentation scheme shown in Chart III.

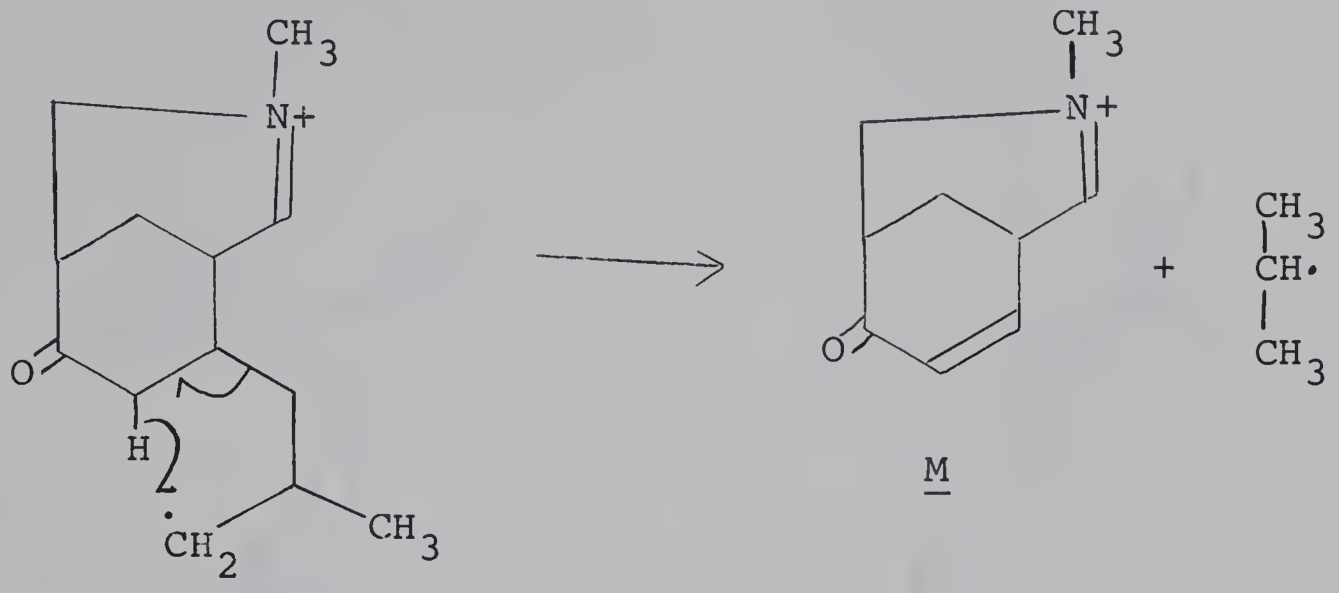
This scheme is similar to the mode of fragmentation that is believed to lead to the loss of C_4H_9 and C_3H_7 in alkaloids of the lycopodine type (55), as shown in the scheme on page 59. In lycopodine the M-C_4H_9 ion is the base peak whereas the M - C_3H_7 ion is less than ten percent of the base peak. In luciduline however, the loss of C_3H_7 (50% of the base peak) is preferred over the loss of C_4H_9 (12% of the base peak) probably due to the bridgehead double bond in the ion D. In fact, the M - C_4H_9 ion may

CHART III


A
B
C
f e d
e
f
d
D m/e 150
+ (CH$_3$)$_2$ĊH
E m/e 164
F
G m/e 164
CH$_3$
N
H
·CH$_2$
O

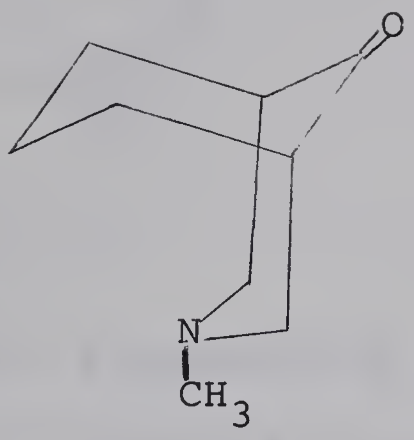

CH3
N
O
CH3
•CH2
H
N
O
CH3
b
CH3
a
N
O
a
b
N
O
(M- C4H9)
N
O
(M- C3H7)

be better represented by M formed as indicated.

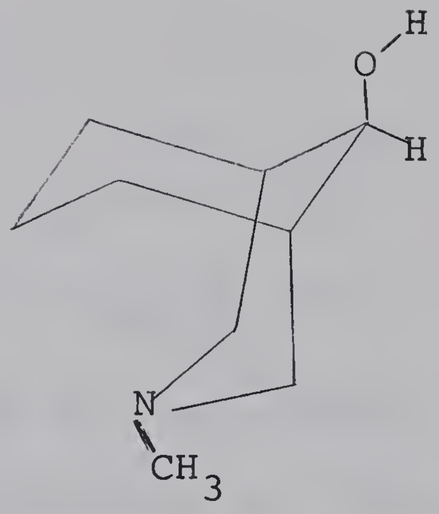

M

Recently, the mass spectra of some derivatives of 3-Azabicyclo- (3.2 .1)-octane and 3-azabicyclo- (3.3.1) -nonane have been reported (71). The aminoketone E and the aminoalcohol F exhibit peaks at m/e 44 with the composition C_2H_6N. It is probable that the m/e 44 fragment in luciduline (A. X & Y = O) and dihydroluciduline (A, X=H, Y=OH) is formed in an analogous way as suggested in the scheme on page 61.

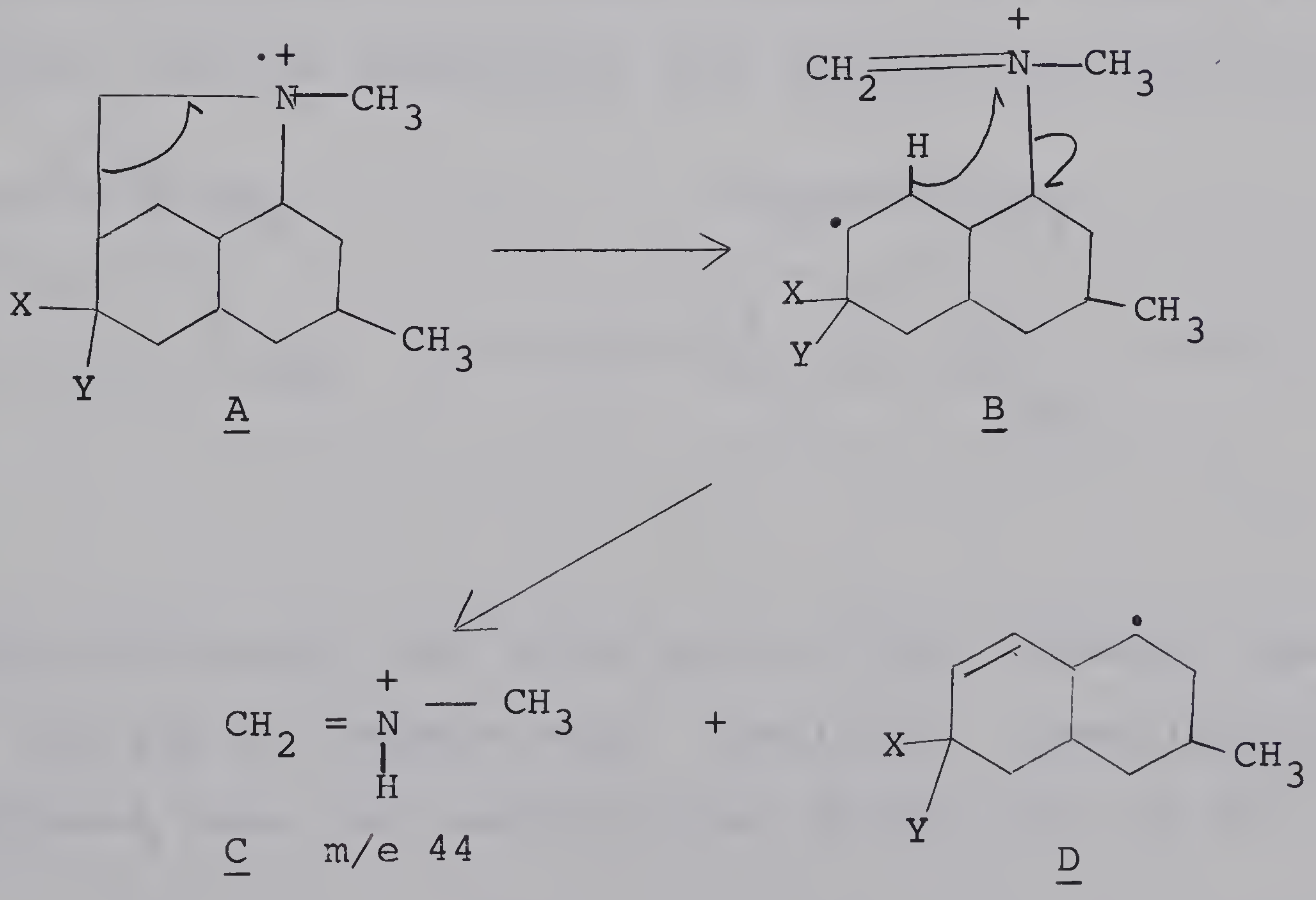
The most prominent peak in the mass spectra of dihydroluciduline, dihydroluciduline -d_1 and dihydroluciduline-d_2 correspond to the loss of a hydroxyl group. Similarly, the O-acyl derivatives that have been prepared exhibit analogous peaks at M-59 and M-199 corresponding to the loss of acetoxyl and p-bromobenzoyloxyl groups respectively.


O
N
CH3

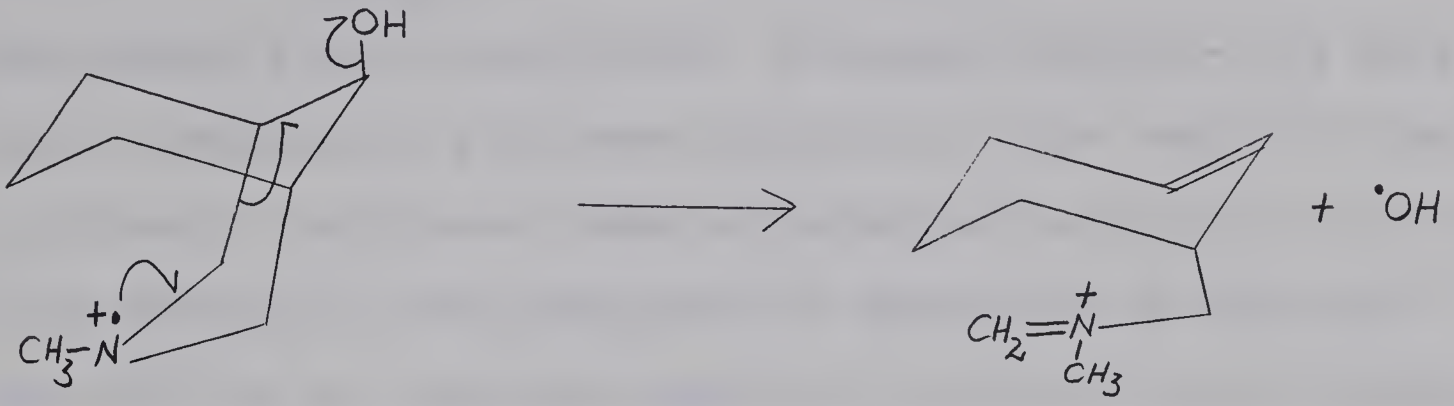
E


H
O
H
N
CH3

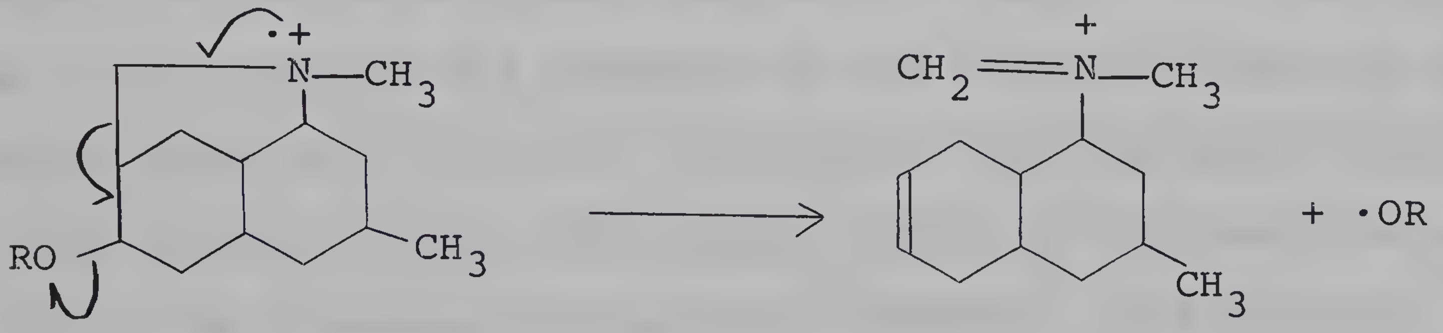
F


·+
N
CH3
X
Y
CH3
A
+
CH2
N
CH3
H
X
Y
CH3
B
CH2 = N+ — CH3
H
C m/e 44
+
X
Y
CH3
D

Some evidence has been adduced to show that the loss of the hydroxyl group in F probably occurs as an electron-impact induced Grob fragmentation as shown below.



This fragmentation is analogous to the normally observed fragmentation of 1,3-aminoalcohols and their derivatives (72). Scheme V illustrates the application of this fragmentation to the interpretation of the mass spectra of dihydroluciduline and its derivatives. This scheme is consistent with the observation that dihydroluciduline-d_1, and



dihydroluciduline-d_2 lose OH to give the most abundant ions at m/e 193 and 194 respectively. Similarly, O-acetyldihydroluciduline-d_2 loses the acetoxyl group forming the ion m/e 194.

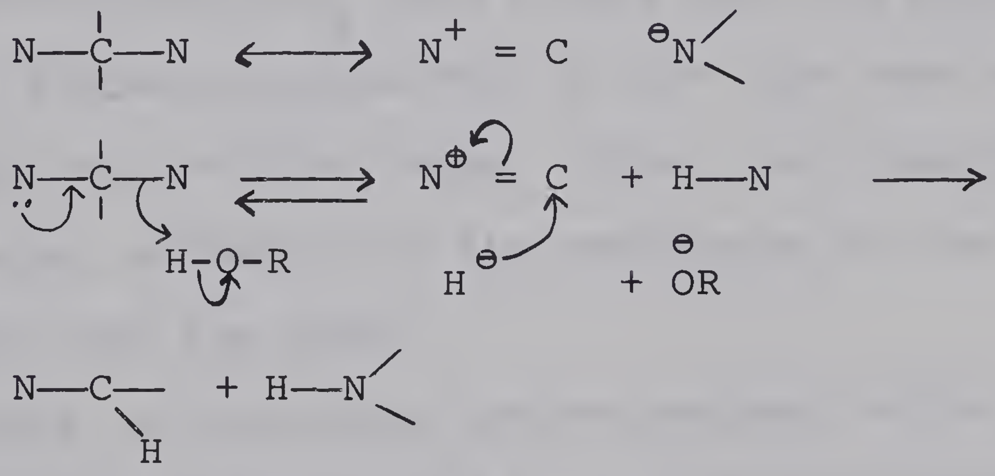
C. Lucidine-A, Lucidine-B and Lycolucine.

Lucidine-A and lucidine-B have the same molecular composition, $C_{30}H_{49}N_3O$, similar spectral characteristics, and undergo similar reactions. Although lucidine-A is more easily obtained as a chromatographically pure compound than lucidine-B, lucidine-A tends to decompose rapidly on standing or in solution. The appearance of absorption at 2780 and 1620 cm^{-1} in the infrared spectra of lucidine-A and lucidine-B in chloroform solution indicates the presence of an N-methyl group (76) and an amide group (77) in the alkaloid. The absence of an amide II band (77) indicates that the amide nitrogen is tertiary. The n.m.r. spectra of lucidine-A and lucidine-B in deuterochloroform provide further information about the nature of these functional groups. The sharp signals at τ7.83 confirms the presence of the N-methyl group and the sharp peaks at τ7.91 and 7.95 suggest that the amide function is an N-acetyl group, the doublet nature of the peaks is probably due to the presence of rotational isomers. In trifluoroacetic acid, the N-methyl peak appears at τ7.30 indicating that the methyl group is attached to a basic nitrogen (53).

The functionality of the third nitrogen in lucidine-B was revealed in the following way. Hydrolysis of lucidine-B with aqueous sulfuric acid gives desacetyllucidine-B, $C_{28}H_{49}N_3$. The infrared spectrum of desacetyllucidine-B in chloroform shows no absorption at 1620 cm^{-1}, but shows an absorption maximum of medium intensity at 1643 cm^{-1} which suggests the presence of an imine group ($>C=N-$) (78). The

assignment of the 1643 cm^{-1} band to an imine function is confirmed by the observation that desacetyllucidine-B is reduced by sodium borohydride or sodium borodeuteride to give desacetyldihydrolucidine-B, $C_{28}H_{51}N_3$, or desacetyldihydrolucidine-B-d$_1$, in which the absorption at 1643 cm^{-1} is no longer apparent.

It is known that compounds which contain an N - C - N group are reduced by sodium borohydride furnishing products which possess a secondary amino group (79) as shown below.



In order to eliminate this possibility we prepared dihydrolucidine-B-d$_1$ by the reduction of lucidine-B with sodium borodeuteride in methanol and compared its mass spectrum with the mass spectra of dihydrolucidine-B and dihydrolucidine-B-d$_2$ prepared by deuterium exchange of dihydrolucidine-B-d$_1$ in methanol-0-d. Some of the most characteristic features in the mass spectral fragmentation of dihydrolucidine-B are the loss of two mass units and the formation of peaks at m/e 275, 273 and 261. It seemed possible that the loss of two mass units was probably associated with the change, $>\mathrm{CH-NH-} \rightarrow\ >\mathrm{C{=}N-}$, in which case the observed mass

spectrometric shifts would be M-4 for a reduced imine group and M-3 for a dideuterated reductively cleaved 1,1-diamine. It was also thought that the appearance of deuterium induced mass spectrometric shifts in the peaks at m/e 275, 273 and 261 might help distinguish between these possibilities. Unfortunately, the high mass range in the mass spectra of dihydrolucidine-B, dihydrolucidine-B-d_1 and dihydrolucidine-B-d_2 did not display the expected differences. Further, dihydrolucidine-B-d_2 gave a mass spectrum which was very similar to dihydrolucidine-B-d_1 in the high mass range as well as in the m/e 260-290 range. Thus, the comparison did not provide any evidence for the preference of one functional group over the other.

In principle, an optically active compound which possesses a functional group which has an absorption with a low molecular absorbance, $\varepsilon \sim 100$, would be expected to display a detectable Cotton effect in the region in which it shows absorption (80). Unconjugated azomethine groups show an $n \rightarrow \pi^*$ transition in the 215-250 mμ region (81,82). Although it was not possible to demonstrate the presence of discernible u.v. absorption in the 215-250 mμ region in lucidine-A and lucidine-B, due to strong end absorption, it seemed likely that lucidine-A, desacetyllucidine-A, lucidine-B and desacetyllucidine-B might exhibit a Cotton effect in this region. The absence of such Cotton effects in the optical rotatory dispersion curves of dihydrolucidine-A and dihydro-

lucidine-B would then constitute evidence in favor of the presence of an azomethine group in lucidine-A and lucidine-B.

The optical rotatory dispersion curves of lucidine-B and desacetyldihydrolucidine-B have been compared with dihydrolucidine-B. Both lucidine-B and desacetyllucidine-B display positive Cotton effect curves with extrema at 260 mμ and 230 mμ whereas dihydrolucidine-B exhibits a plain curve. Similarly, lucidine-A and desacetyllucidine-A display Cotton effect curves in the 300-200 mμ region whereas dihydrolucidine-A shows only a plain curve in its optical rotatory dispersion curve. These results strongly suggest the presence of an imine group in lucidine-A and lucidine-B.

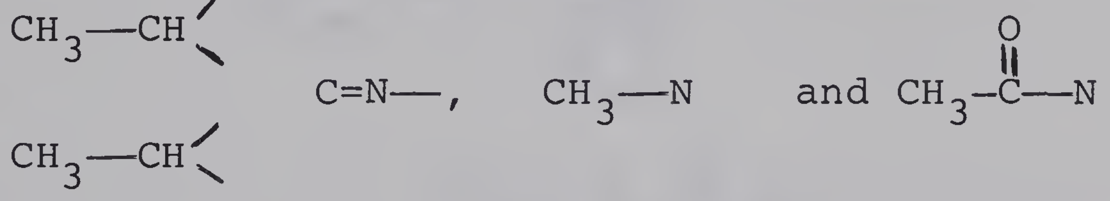
The chemical shift of the N-methyl group in both deuterochloroform and trifluoroacetic acid, and the absorption at 2780 cm^{-1} clearly indicate that the acetyl group is not bonded to the nitrogen carrying the methyl group, but do not exclude the possibility that the methyl group is bonded to the imine nitrogen. This possibility is unlikely since it is found that reduction of the imine function with sodium borohydride and acetylation of the newly formed secondary amine group gives a compound, N-acetyldihydrollucidine-B, $C_{32}H_{53}N_3O_2$, in which the N-methyl group resonates around τ7.8 and not at τ7.0 to 7.2 as might be expected for an N-methyl group attached to an amide nitrogen.

In addition to the N-methyl and N-acetyl groups, the n.m.r. spectra of lucidine-A and lucidine-B show complex multiplets

in the τ8.9-9.2 region indicating the presence of C-methyl groups.

The number of C-methyl groups is difficult to determine from the n.m.r. spectrum but the appearance of two doublets in the C-methyl region in lucidine-B suggests the presence of two secondary methyl groups. Since Kuhn-Roth oxidation of desacetyldihydrolucidine-B gives rise to 1.12 moles of acetic acid, lucidine-B probably possesses two secondary methyl groups.

The above data indicates the presence of the following functional groups in lucidine-A and lucidine-B:

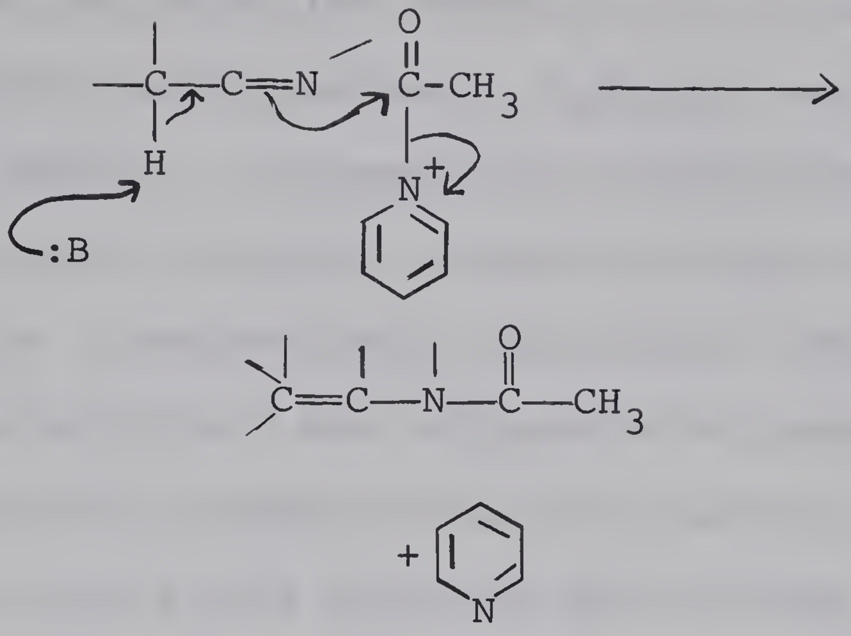


$$CH_3{-}CH\langle \quad CH_3{-}CH\langle \quad C{=}N{-}, \quad CH_3{-}N \quad \text{and} \quad CH_3{-}\overset{O}{\overset{\|}{C}}{-}N$$

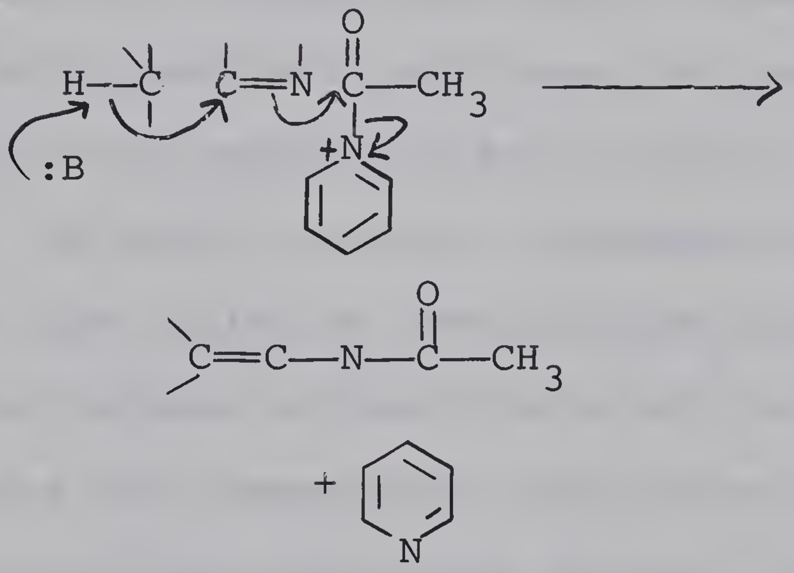
Preliminary experiments on a mixture of lucidine-A and lucidine-B indicated that acetylation of the desacetyl derivatives does not give a mixture of lucidine-A and lucidine-B. Lucidine-A and lucidine-B themselves appeared to be acetylated when treated with acetic anhydride in pyridine at room temperature. When this experiment was carried out on pure lucidine-B, it was found that N-acetyllucidine-B, molecular weight 509, $C_{32}H_{51}N_3O_2$, was formed. Since dihydrolucidine-B reacts with only one equivalent of acetic anhydride to form N-acetyldihydrolucidine-B, molecular weight 511, $C_{32}H_{53}N_3O_2$, the acetylation of lucidine-B may involve the C=N- group as outlined in Schemes

I and II below:

SCHEME I



SCHEME II



The reaction shown in Scheme II is analogous to the Mannich reaction (83) whereas the one shown in Scheme I constitutes the normal mode of reaction between imines and acetylating agents (84). The apparent absence of activating groups in lucidine-B suggests that the acetylation is not likely to occur as indicated in Scheme II.

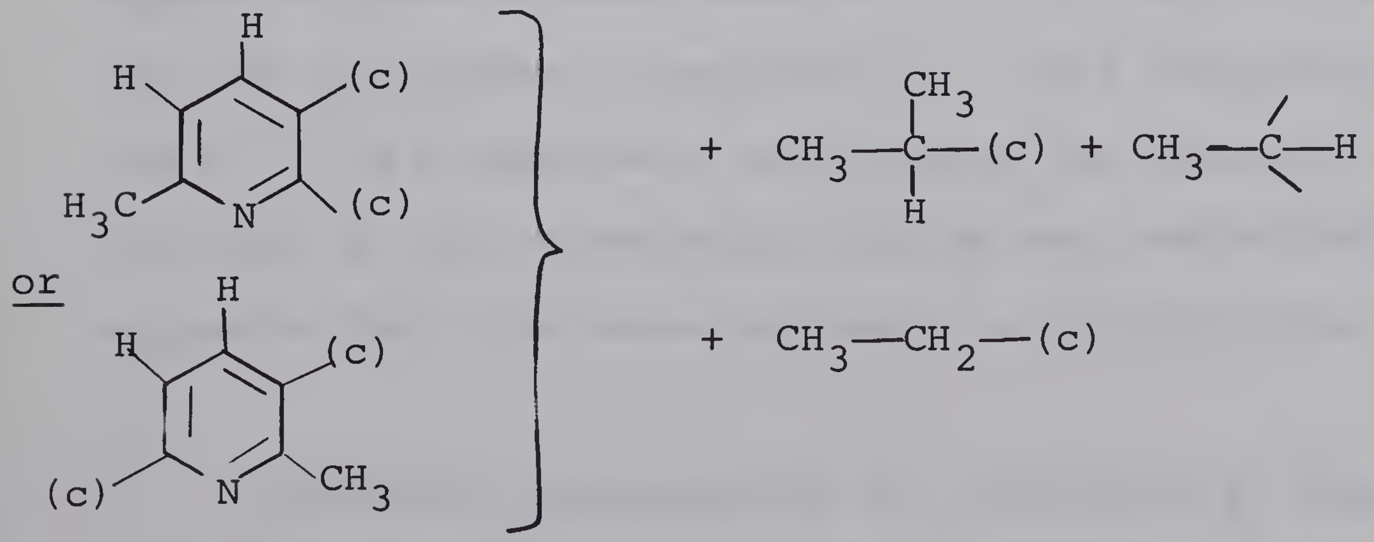
The reduction of dihydrolucidine-B with lithium aluminium hydride (61) gave tetrahydrodeoxylucidine-B as a

white amorphous solid which analysed for $C_{30}H_{53}N_3$. Its n.m.r. spectrum indicated the presence of an N-CH_2-CH_3 group. Tetrahydrodeoxylucidine-A, $C_{30}H_{53}N_3$, was prepared in a similar fashion. On acetylation, tetrahydrodeoxylucidine-B, gives an N-acetyl compound, molecular weight 497, indicating the formation of a mono-N-acetyl derivative. When N-acetyltetrahydrodeoxylucidine-B was refluxed with iodomethane in methanol, methiodide formation was not observed. Similarly disappointing results were obtained when attempts were made to derivatize lucidine-B, dihydrolucidine-B, desacetyldihydrolucidine-B, desacetyllucidine-B and N-acetyldihydrolucidine-B by salt formation with perchloric acid, picric acid, hydrogen bromide, and methyl iodide. Compounds in the lucidine-A series also failed to furnish crystalline derivatives.

The molecular formula of lucidine-A and lucidine-B, $C_{30}H_{49}N_3O$, indicates the presence of eight sites of unsaturation. The N-acetyl group and the imine group account for two sites of unsaturation. Since nuclear magnetic resonance and infrared spectra of lucidine-A and lucidine-B and their derivatives do not show any indication of any other unsaturation, lucidine-A and lucidine-B are therefore hexacyclic compounds. An experiment on dihydrolucidine-B showed that it is recovered unchanged (i.r. and t.l.c.) after shaking with hydrogen at 50 atmospheres for 20 hours. The nature of these results do not, however, exclude the possibility of the presence of a hindered non-conjugated tetrasubstituted double bond in

Column chromatography of the weakly basic dehydrogenation products on alumina and gas-liquid chromatography of the fraction eluted with a hexane-benzene (1:1) mixture led to the isolation of two similar compounds, designated GC-17 and GC-18. GC-17 and GC-18 have the same molecular formula, $C_{17}H_{25}N$, and similar spectral characteristics (UV, n.m.r. and mass spectra). The ultraviolet spectra of GC-17 and GC-18 indicate that they are 2,3,6-trisubstituted pyridines (75). The n.m.r. spectrum of GC-18 (Fig. 30) shows two aromatic protons as an AB quartet (J=8 cps) centered at τ2.91, a methyl singlet at τ7.45, an isopropyl group at τ8.58 as a doublet (J=6 cps) and C-methyl multiplets in the τ9.00-9.10 region. Irradiation experiments indicate that the isopropyl group is coupled to the τ7.01 region and that the C-methyl multiplets are coupled to τ8.54 and 8.02. The C-methyl multiplets appear to consist of a methyl triplet and an ethyl doublet. The methyl group at τ7.45 is likely to be bonded to the α-position of the pyridine ring (89).

These spectral characteristics suggest that GC-18 has one of the partial structures indicated below:



lucidine-A and lucidine-B.

In addition to lucidine-A and lucidine-B, we have isolated lycolucine, $C_{30}H_{43}N_3O$, as a white crystalline solid, m.p. 198-200°. It resembles lucidine-A and lucidine-B in that it has both an N-methyl and N-acetyl group as indicated by absorption at 2775 cm^{-1} (NCH_3) and 1651 cm^{-1} ($N-CO-CH_3$) in its infrared spectrum in carbon tetrachloride. The presence of peaks at τ7.88 and 7.93 in its n.m.r. spectrum in deuterochloroform and the fact that hydrolysis with dilute sulfuric acid removes the N-acetyl group, forming desacetyl lycolucine, $C_{28}H_{41}N_3$, also supports this assumption. The n.m.r. spectra of both lycolucine and desacetyllycolucine indicate the presence of two C-methyl groups in the τ9.0-9.2 region.

Lycolucine differs from lucidine-A and lucidine-B in that it has three more unsaturations. Its ultraviolet spectrum exhibits an unusual chromophore with maxima at 217(ε=14,530), 261(ε=8,800), 272(ε=7,820) and 300(ε=10,200) mμ. When a few drops of dilute hydrochloric acid are added, the absorption maxima at 217, 261 and 272 mμ appear as less intense bands at 210, 258 and 268 mμ while the 300 mμ maximum is replaced by a more intense band at 320 mμ. This experiment establishes the presence of nitrogen in the chromophoric system and, tentatively, suggests that lycolucine possesses a vinylpyridine system.

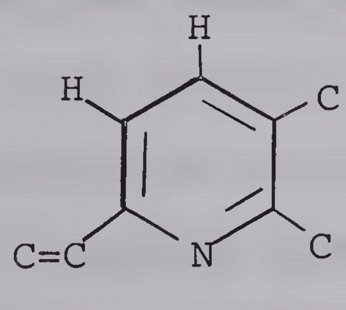
Catalytic hydrogenation of lycolucine at room

temperature and atmospheric pressure gives dihydrolycolucine, a compound whose molecular weight corresponds to the formula $C_{30}H_{45}N_3O$. The ultraviolet spectrum of dihydrolycolucine has maxima at 272 mμ and 278 mμ. On acidification, the 272 mμ undergoes a bathochromic shift to 278 mμ. Andon, Cox and Herington (75) have derived the following emperical relationships between absorption maxima and the position of alkyl substituents in pyridines:

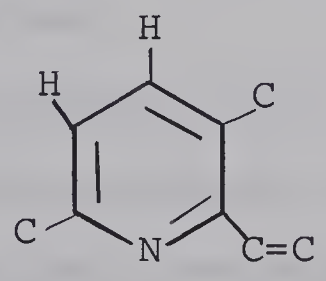
$$\lambda_{max} = 257 + 5a + 6b - 2c, \quad \text{in alkaline solution}$$

$$\lambda_{max} = 256 + 7a + 6.5b - 3c, \quad \text{in acid solution}$$

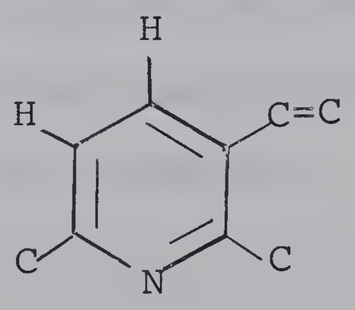
where a, b and c are the number of α, β, and γ substituents respectively. The spectral data strongly suggest that dihydrolycolucine has a 2,3,6-trisubstituted pyridine system since it has absorption maxima at 275 mμ in neutral solution and at 280 mμ in acid solution (λ_{max} calculated 273 and 276.5 mμ respectively). The ultraviolet spectra therefore suggest the partial structures I, II and III for lycolucine.



I



II

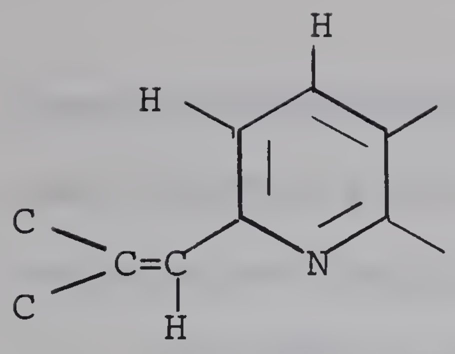


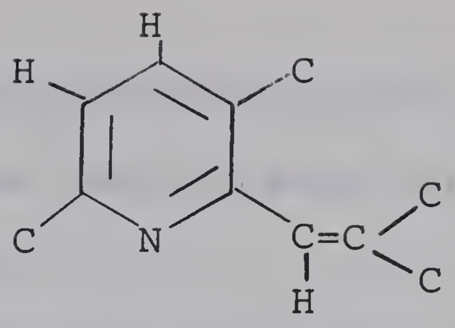
III

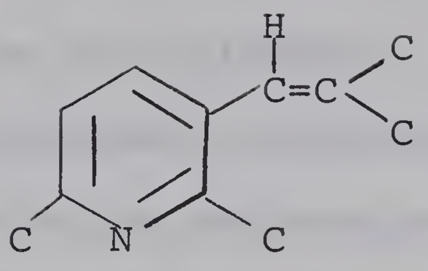
The n.m.r. spectrum of lycolucine (Fig. 7) in deuterochloroform shows the presence of three protons in the aromatic region centered at τ2.70, 3.20 and 3.43. Although all the signals are split, the signals at τ2.70 and 3.20 constitute an AB quartet, J=8 cps, which we assign to the β- and γ-pyridine protons. The chemical shift data and splittings reported for β- and γ-protons in pyridines (85) are similar to the values obtained for lycolucine. The one proton signal at τ3.43 is assigned to a vinyl proton.

The n.m.r. spectrum of desacetyllycolucine amply supports the above assignments. The protons in the aromatic region comprise a well-defined AB quartet, J=8 cps, centered at τ3.05 and a broad one proton singlet at τ3.40. Thus it seems probable that the further splitting of the aromatic region in lycolucine arises from the rotomeric or other effect of the N-acetyl group. Since the ultraviolet spectrum of desacetyllycolucine is practically identical with that of lycolucine it is unlikely that any rearrangement occurred during the formation of desacetyllycolucine.

The chemical shifts of the vinyl protons in 2-vinylpyridine and 4-vinylpyridine have been reported (86). The observed chemical shifts indicate that lycolucine possesses a β-disubstituted vinylpyridine grouping as shown in partial structures IV, V and VI.


IV.


V.


VI.

The similarities between the molecular formulas of lycolucine and lucidine-A indicated that octahydrodesacetyllycolucine might be structurally similar to desacetyldihydrolucidine-A. Hydrolysis of dihydrolycolucine gave desacetyldihydrolycolucine (molecular weight, 421), and reduction of desacetyldihydrolycolucine with sodium in ethanol gave octahydrodesacetyllycolucines (molecular weight, 427) as an amorphous solid which shows several components on t.l.c. One of the minor components has the same R_f value as desacetyldihydrolucidine-A.

The lack of complete identity is certainly not unexpected since the reduction of the 2,3,6-trisubstituted pyridine system by dissolving metals is expected to produce stereoisomeric piperidines. Although t.l.c. comparison indicated only some similarity, the mass spectral fragmentation pattern of octahydrodesacetyllycolucine is strikingly similar to the fragmentation pattern of the

desacetyldihydrolucidines. The close similarity of the mass spectra indicates that lycolucine has the same carbon skeleton as lucidine-A and lucidine-B.

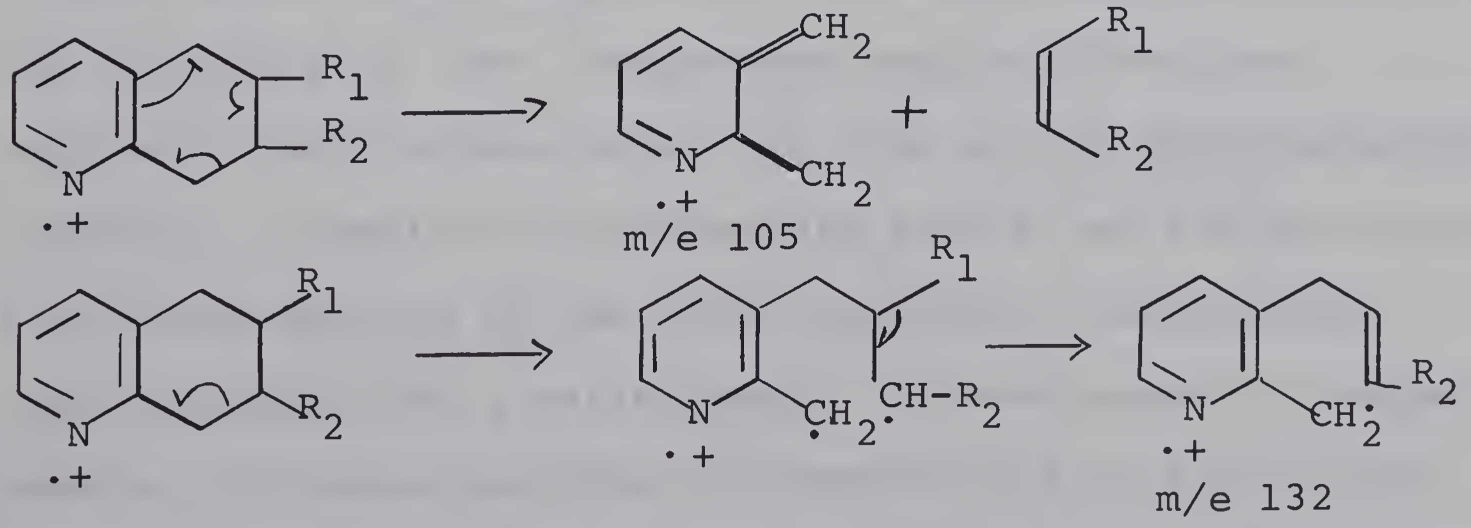
In order to gain further insight into the nature of the carbon skeleton in lucidine-A and lucidine-B, we have subjected the lucidines, dihydrolucidine-B, and dihydrolucidine-A to selenium dehydrogenation under a variety of conditions. Most of the dehydrogenation experiments were done on dihydrolucidine-B, but we have shown that dihydrolucidine-A and the lucidines give the same major products as dihydrolucidine-B on dehydrogenation.

The mixture of dehydrogenation products was first separated into non-basic, weakly basic, and strongly basic fractions using the procedure described in the experimental section of this thesis. A combination of column chromatography and gas-liquid chromatography on the non-basic fraction led to the isolation of small quantities of a hydrocarbon, $C_{12}H_{10}$ which has been shown to be diphenyl (U.V., mass spectrum, and g.l.c. retention time). The strongly basic fraction was partially separated by means of counter-current distribution over nine funnels at pH 4.5 using potassium dihydrogen phosphate as the stationary phase and methylene chloride as the moving phase. The fractions obtained from the counter-current distribution were then further analysed by gas-liquid chromatography. Separation of the components by gas-liquid chromatography has led to the isolation of three C_{10}

nitrogen-containing compounds, I($C_{10}H_9N$), II ($C_{10}H_{13}N$) and III ($C_{10}H_{19}N$). A comparison of the retention times of the seven monomethylquinolines with compound I showed that I was either 6 or 7-methylquinoline. Compound I has been identified as 7-methylquinoline. The infrared, ultraviolet and mass spectra of compound I are identical with spectra of authentic 7-methylquinoline (87).

The application of the relationships derived by Andon, Cox and Herington (75) to the ultraviolet spectrum of compound II indicated that it is a 2,3- or 2,5-disubstituted pyridine. Its molecular composition, $C_{10}H_{13}N$, and mass spectral fragmentation (Fig. 27) suggest that it is 6- or 7-methyl-5,6,7,8-tetrahydroquinoline.

Although the mass spectral fragmentation of compound II is consistent with both 6- and 7-methyl-5,6,7,8-tetrahydroquinoline, the isolation of 7-methylquinoline strongly suggests that compound II is 7-methyl, 5,6,7,8-tetrahydroquinoline. The interpretation of its mass spectrum is summarized in the chart below.



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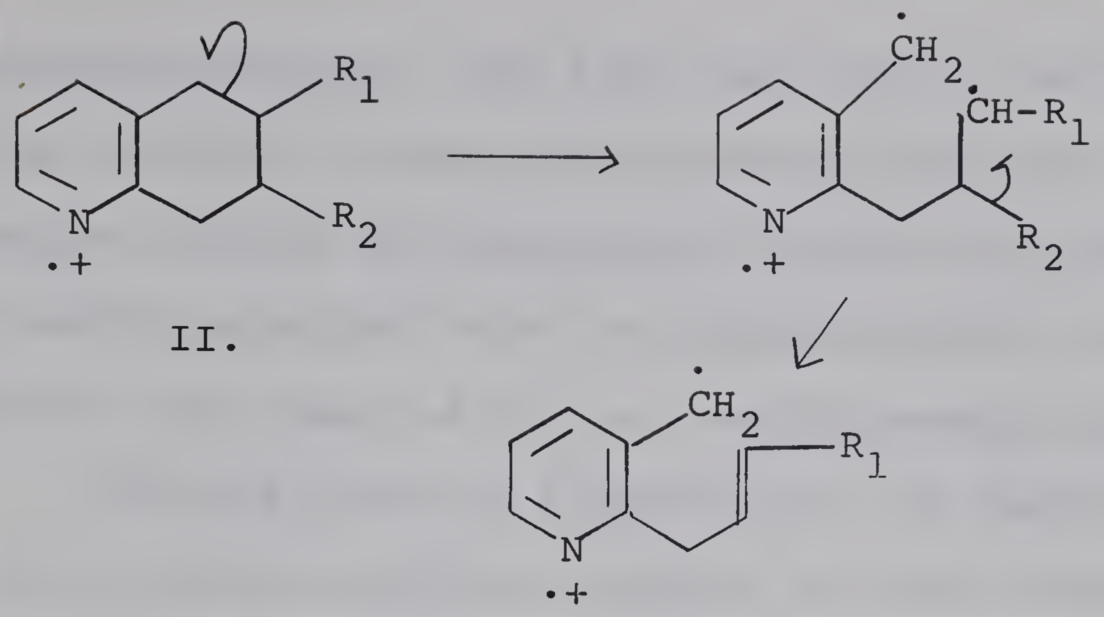


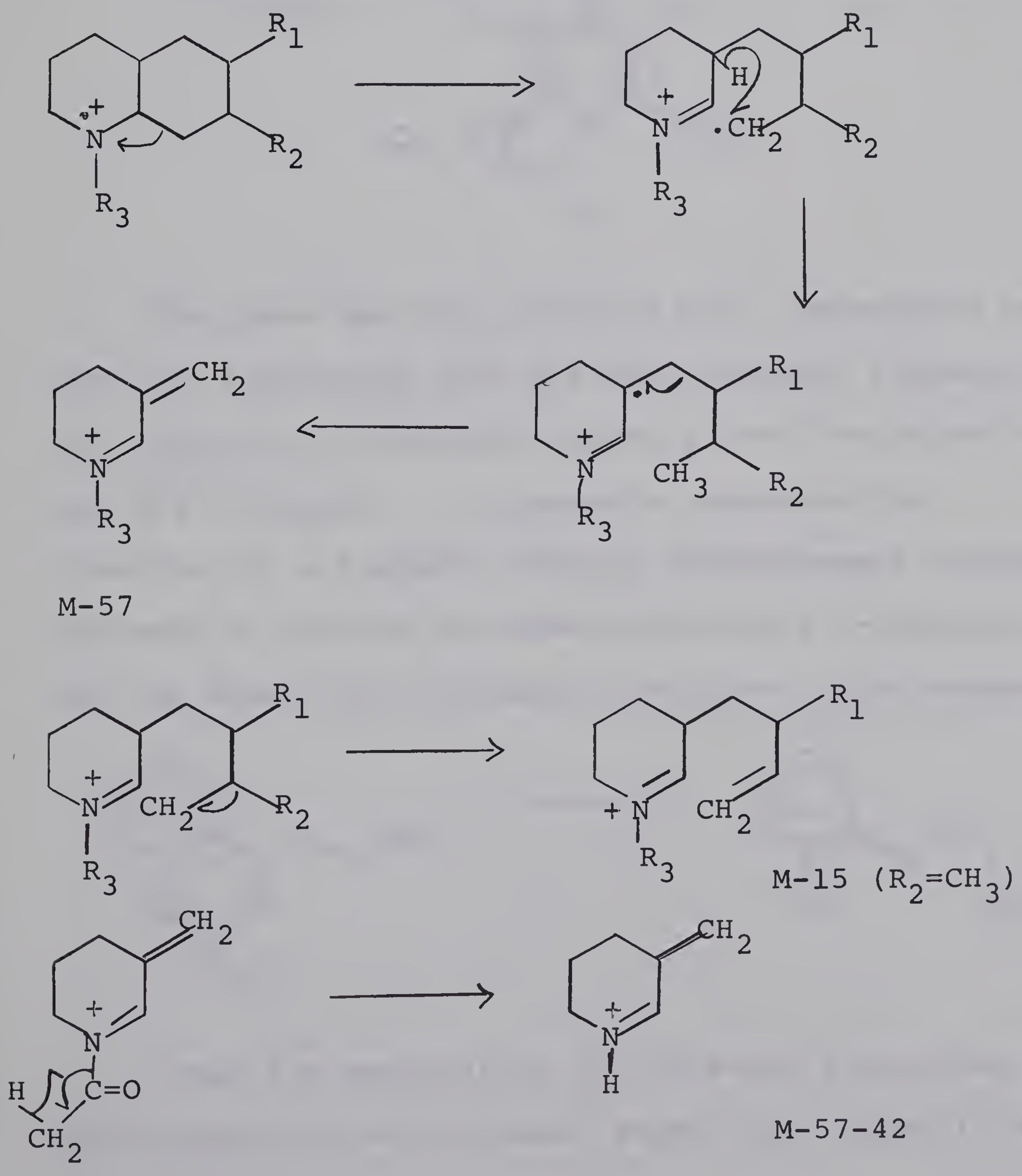
II.

Compound III ($C_{10}H_{19}N$) shows no ultraviolet absorption above 215 mμ and reacts with acetic anhydride at room temperature to form an N-acetyl derivative. The mass spectra of compound III and its N-acetylderivative (IV) suggest that it is 6- or 7-methyldecahydroquinoline. It will be seen from the fragmentation scheme below that it is difficult to distinguish between 6- and 7-methylquinoline from the gross mass spectral features.

In order to distinguish between these two alternatives, 7-methyldecahydroquinoline was prepared by the catalytic reduction of 7-methylquinoline (88). Gas-liquid chromatography showed that the synthetic 7-methyldecahydroquinoline was a mixture of two components and only the minor component had the same retention time as the dehydrogenation product. In spite of considerable effort, we did not collect sufficient amounts of the minor synthetic 7-methyldecahydroquinoline for a satisfactory infrared spectral comparison. However, the mass spectrum of compound III is practically identical with the mass spectrum of the mixture of 7-methyl-

decahydroquinolines. The fact that one of the components of the synthetic 7-methyldecahydroquinoline has the same retention time as the degradation product and the isolation of 7-methylquinoline from the dehydrogenation mixture indicate that compound III is 7-methyldecahydroquinoline.

The mass spectral fragmentation of compounds III and IV is then explained quite readily as shown below.



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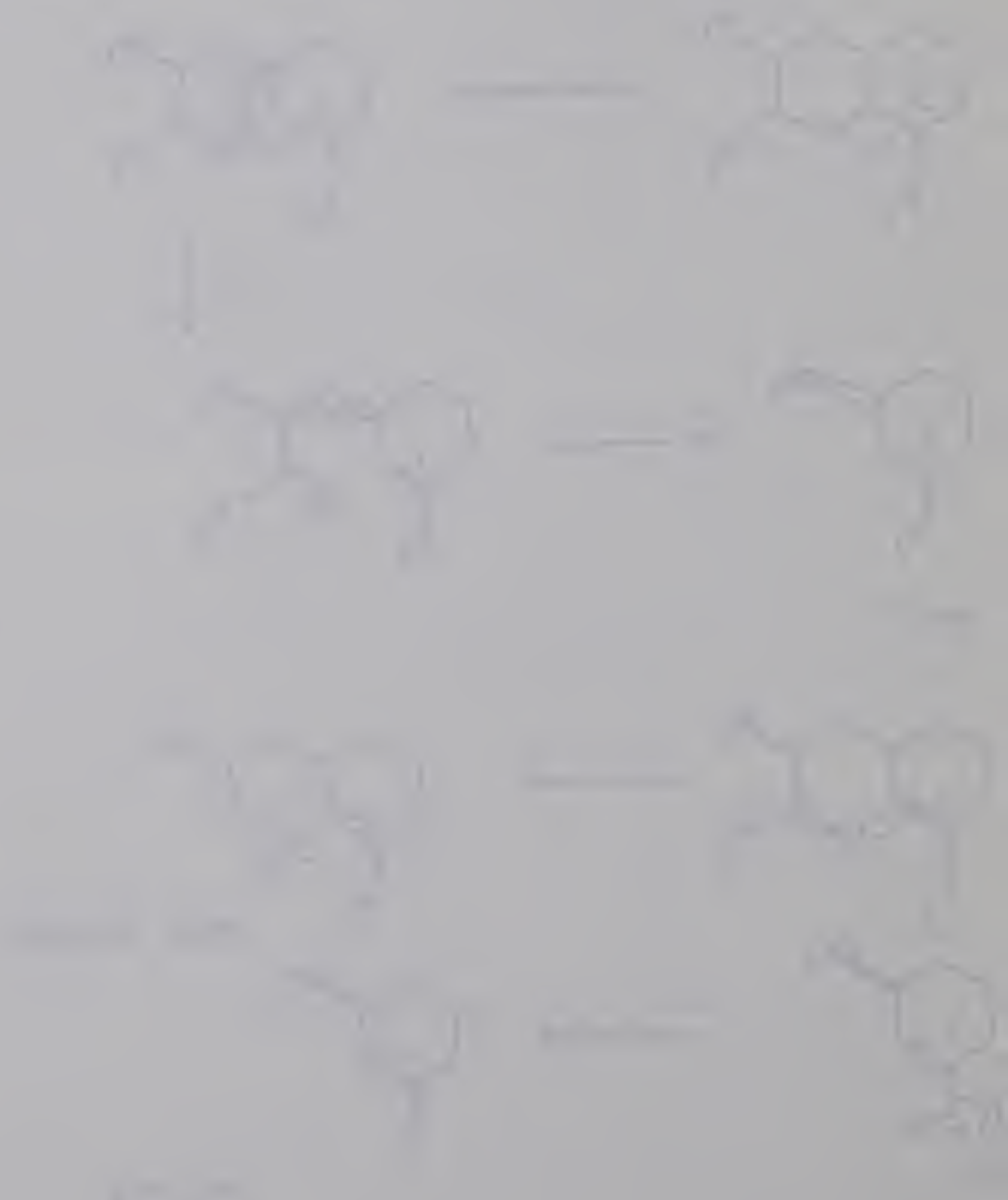
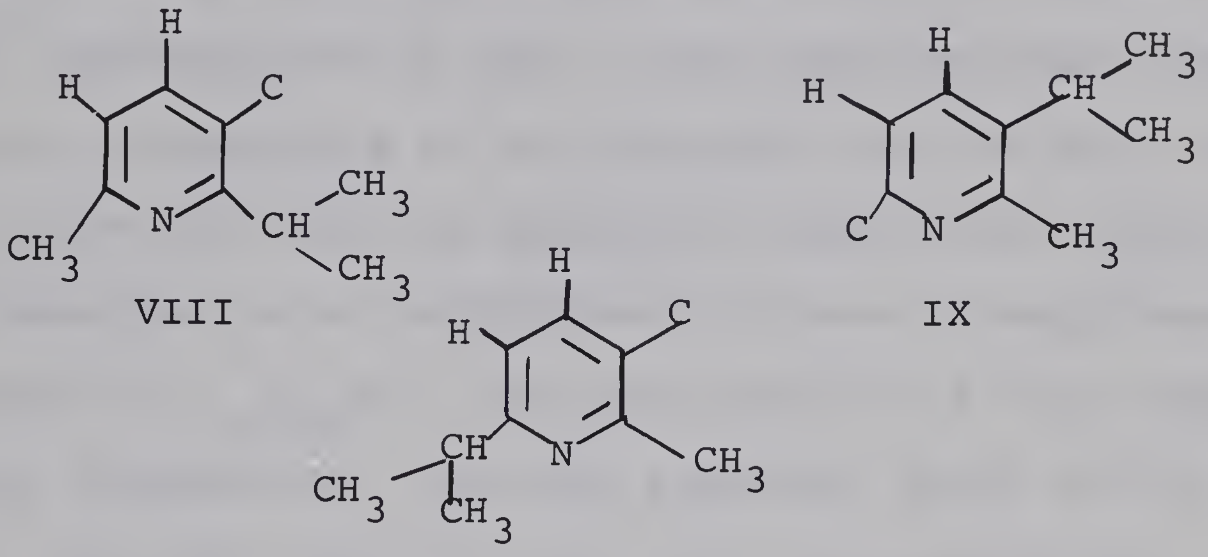


The chemical shifts of the isopropyl group and the proton to which it is coupled suggest that it could quite probably be attached to the aromatic ring, thus leading to the partial structures VIII, IX and X as shown below.

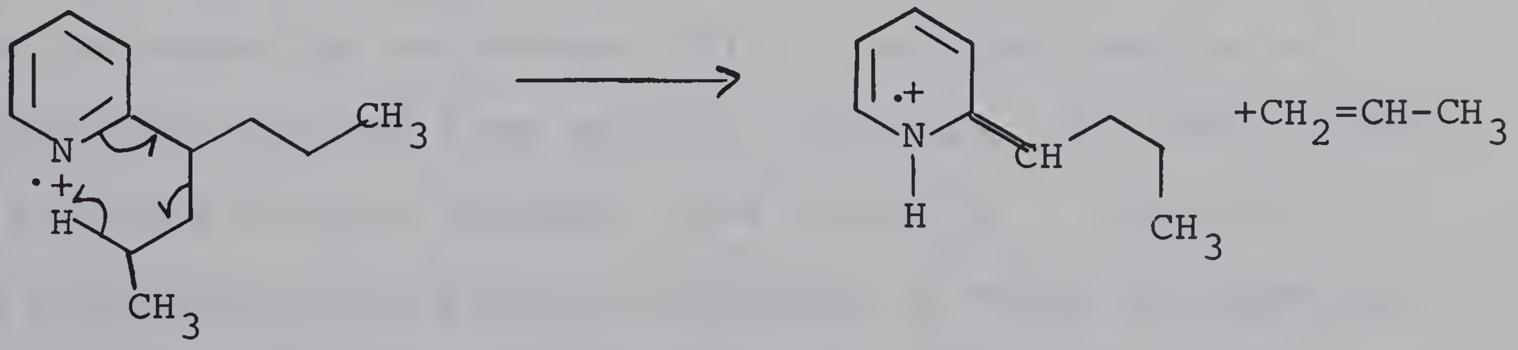


VIII

IX

X

The mass spectrum of GC-18 has a metastable peak at m/e 75.0 indicating that the most abundant fragment, m/e 135 ($C_9H_{13}N$), is obtained directly from the molecular ion m/e 243 ($C_{17}H_{25}N$). In aromatic compounds the formation of a fragment through rearrangement processes is believed to involve the abstraction of a γ-hydrogen atom (90) as shown for 2-(4-heptyl)pyridine in the scheme below.



When the application of the basic principles of mass spectrometry to GC-18 itself seemed not to facilitate the

efforts to elucidate the structure of GC-18, we decided to utilize the mass spectra of its perhydroderivatives. The reduction of GC-18 with a large excess of sodium in ethanol gave the hexahydroderivative of GC-18 in low yield. Hexahydro-GC-18 gave a mass spectrum which showed no peak corresponding to the molecular ion but had a base peak at m/e 124 with the molecular composition, $C_8H_{14}N$.

Acetylation of hexahydro-GC-18 gave N-acetylhexahydro-GC-18, $C_{19}H_{33}NO$. The mass spectrum of this compound is very informative. The most prominent peaks at m/e 291 (18%), 166(100%) and 124 (41%) have the compositions $C_{19}H_{33}NO$, $C_{10}H_{16}NO$ and $C_8H_{14}N$ as shown by accurate mass measurements. The presence of metastable peaks at m/e 94.6 and 92.8 indicate that the following fragmentation sequence occurs:

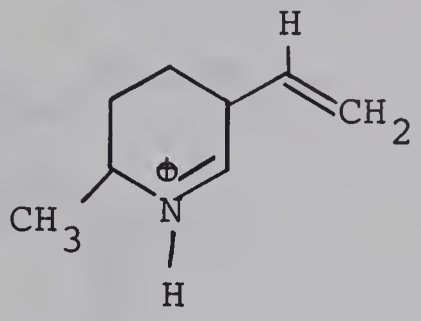
$$[C_{19}H_{33}NO]^+ \rightarrow [C_{10}H_{16}NO]^+$$

$$[C_{10}H_{16}NO]^+ \rightarrow [C_8H_{14}N]^+$$

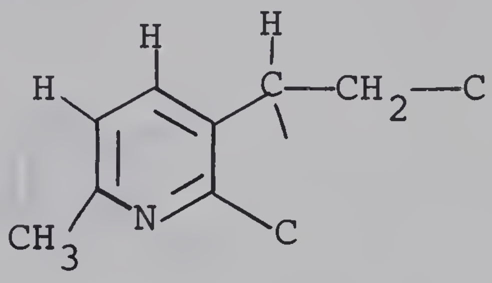
The most abundant ions in piperidines arise from the ion formed by α-cleavage (91). The mass spectra of piperidines derived from partial structures VIII and X would be expected to have abundant ions at M-C_3H_7. Similarly, the piperidine from partial structure IX would be expected to have a prominent C_9 fragment. The absence of these types of ions in the mass spectrum of N-acetylperhydro-GC-18 provide strong evidence against partial structures VIII, IX

and X.

If it is assumed, by analogy to the decahydroquinolines (see above), that the m/e 124 fragment has the structure XI, then the pyridine GC-18 has the partial structure XII.



XI

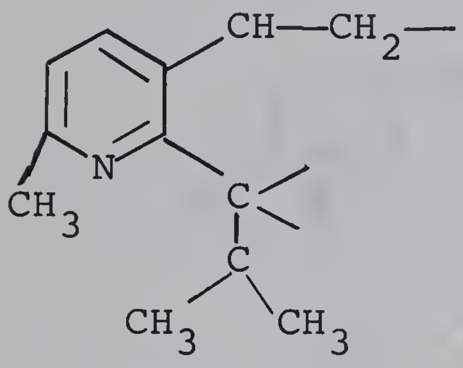


XII

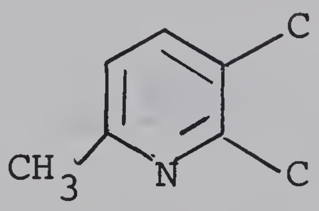
In the course of the investigation of GC-18 we made attempts, albeit thus far unsuccessful, to derive structures for GC-18 which would be consistent with all the observed spectral properties. A few of the attempts to develop suitable working hypotheses are summarized below.

The n.m.r. and ultraviolet spectral data suggested the partial structures I-IV in the chart below. Since the mass spectra of GC-18 and N-acetylhexahydro-GC-18 indicate that the isopropyl group is not attached directly to the aromatic ring, it seemed possible that its attachment to the α-carbon atom as in structure V might result in an isopropyl group with the same chemical shifts as obtained for GC-18. The presence of a β Ar-CH-CH_2 group is inferred from the appearance of the m/e 124 peak in N-acetylhexahydro-GC-18. Structure V plus the secondary methyl and the ethyl group

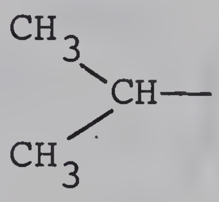
account for all but two of the carbon atoms in $C_{17}H_{25}N$. Since there is no evidence for a double bond* the degradation product is assumed to be tricyclic and one can arrive at structures such as VI ($R_1=CH_3$; $R_2=CH_2CH_3$) and VII (VI $R_1 = CH_2CH_3$; $R_2=CH_3$).



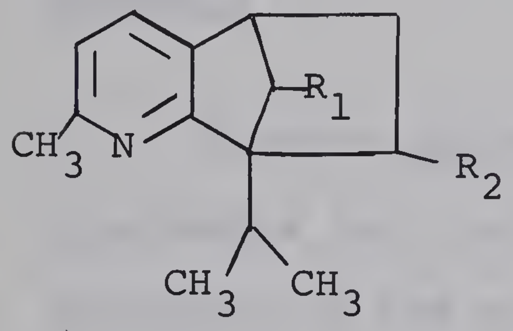
V



I



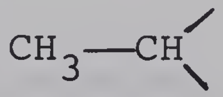
II



VI

CH_3-CH_2-

IV



III

The most attractive feature about structure VI or VII is that it readily rationalizes the mass spectral fragmentation of N-acetylhexahydro-GC-18 (VIII) as shown in Scheme A. Structures VI and VII however do not easily lead to an explanation for the mass spectrum of GC-18 itself.

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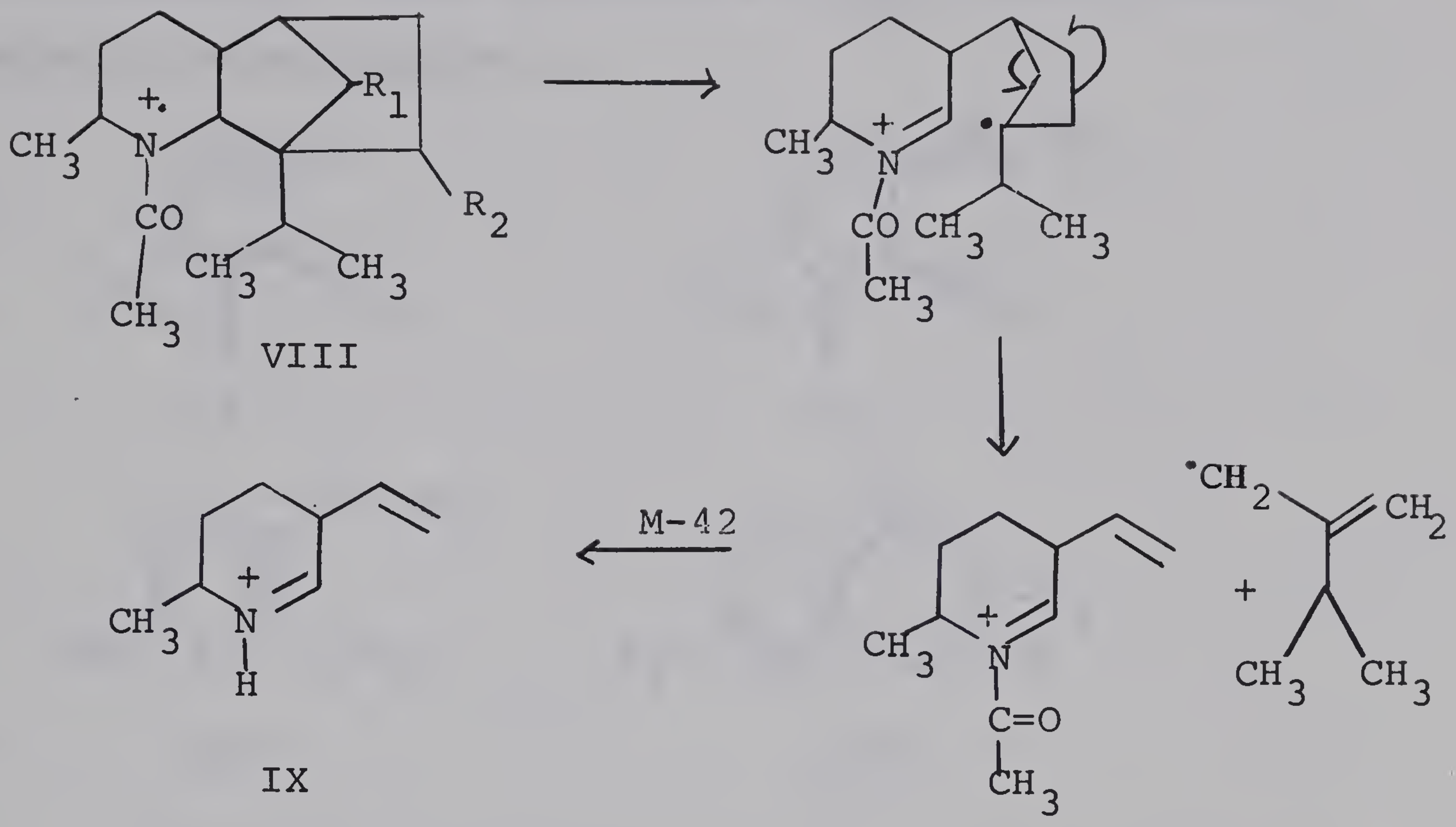
* GC-18 was recovered unchanged (mass spectrum) when subjected to catalytic hydrogenation over Adam's catalyst at atmospheric pressure for three days.



Scheme A

Another approach to the elaboration of a working hypothesis for the structure of GC-18 starts with the structure of the m/e 124 fragment (IX). Structure IX suggests the possibility that the m/e 135 fragment in the mass spectrum of GC-18 might have the partial structure X and hence the structure XI. If one assumes a minimum number of hydrogen transfers in the formation of XI, then GC-18 can be visualized as possessing the partial structure XII. Since n.m.r. spectra of GC-18 suggest the presence of CH_3CH , $(CH_3)_2CH-$ and CH_3CH_2- groups, one can arrive at structures of type XIII (R_1, R_2 and R_3 are CH_3, $(CH_3)_2CH$, or CH_3CH_2). The hindered nature of the tetrasubstituted double bond is not inconsistent with the fact that GC-18 is recovered unchanged on attempted

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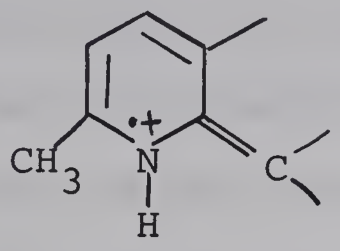
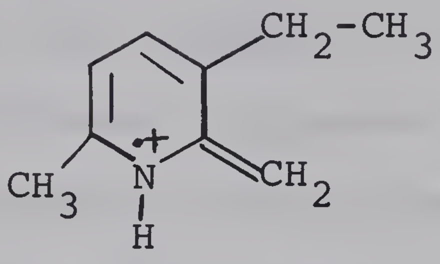
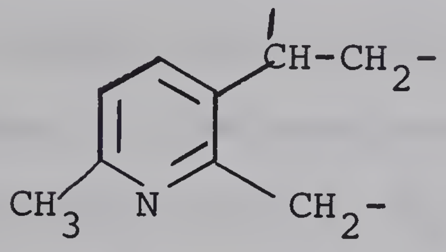
hydrogenation. These structures do not however easily explain the mass spectra.



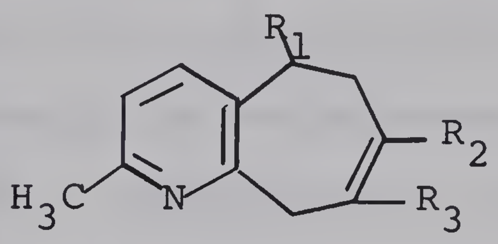
X



XI



XII

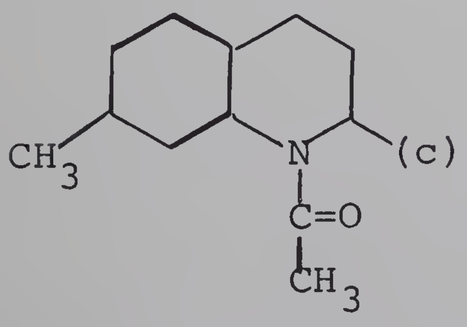


XIII

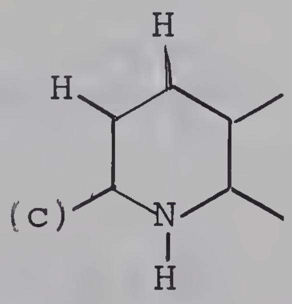
In spite of the inconclusive nature of the dehydrogenation experiments, it appears that the C_{10} compounds such as 7-methylquinoline and the C_{17} compounds such as GC-18 constitute the two main parts of the structures of lucidine-A and lucidine-B. The fact that the major basic dehydrogenation product of lycolucine is identical with the pyridine, $C_{10}H_{13}N$, obtained by the dehydrogenation of the lucidines and that GC-17 and GC-18 are also formed from lycolucine, demonstrates that lucidine-A and lucidine-B have the same carbon skeleton as lycolucine.

The formation of 7-methyldecahydroquinoline even under very mild conditions suggests that lucidine-A, lucidine-B and lycolucine possess a skeleton which readily gives 7-methyldecahydroquinoline. This view is also in agreement with the observed formation of 7-methyl-5,6,7,8-tetrahydroquinoline on pyrolysis of dihydrolucidine-A.

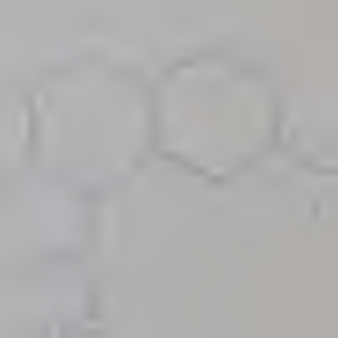
The similarity between the pyrolysis products and mass spectral fragments has been reported (92). It seemed possible that the $C_{10}H_{16}N$ (m/e 150) fragment in the mass spectrum of tetrahydrodeoxylucidine-B originates from the same part of the molecule of lucidine-B that gives rise to the 7-methyldecahydroquinoline. Such speculation leads to a further possibility that the $C_{17}H_{29}N_2$ (m/e 261) fragment in the mass spectrum of tetrahydrodeoxylucidine-B is related to GC-18 ($C_{17}H_{25}N$). Comparison of the mass spectra of dihydrolucidine-B and tetrahydrodeoxylucidine-B and tetrahydrodeoxylucidine-B-d_3 (figures 18, 22 and 23) shows that the most notable effect of the replacement of an acetyl group by an ethyl group is the formation of a prominent ion at m/e 180 ($C_{12}H_{22}N$) while the m/e 261 ($C_{17}H_{29}N_2$) fragment remains prominent. A combination of all these observations have led us to speculate that for the dihydrolucidines, the two partial structures, I and II, are probably joined via an N—C—CH_2—C—N fragment as indicated in partial structures III and IV in the chart below:

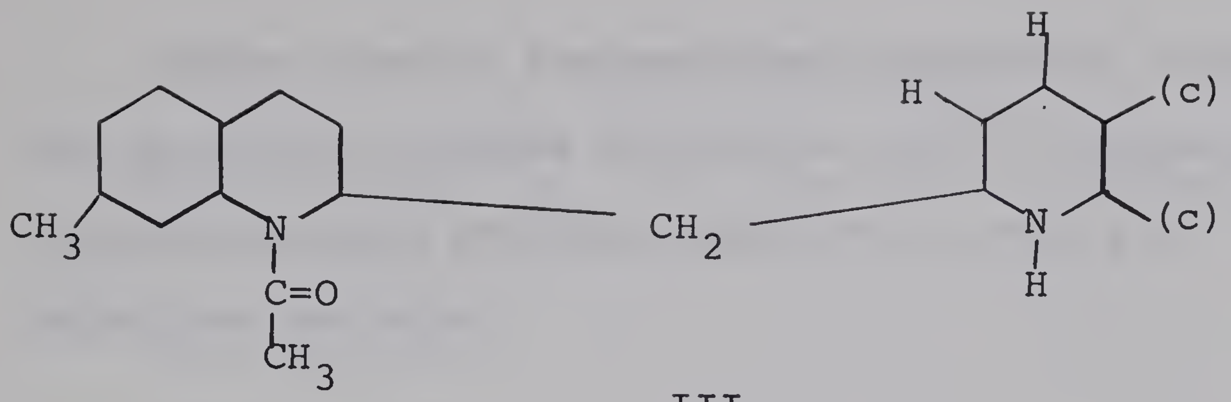


I.

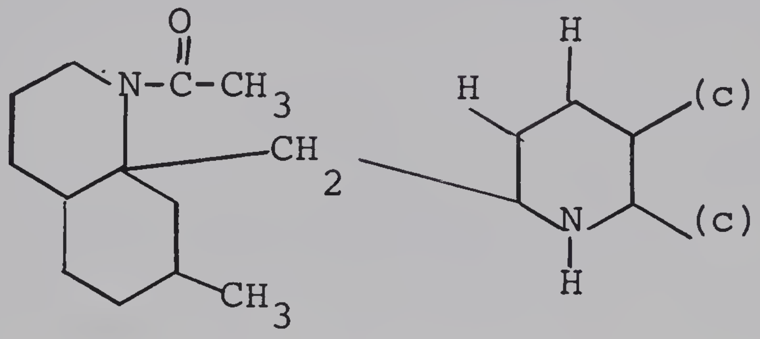


II.



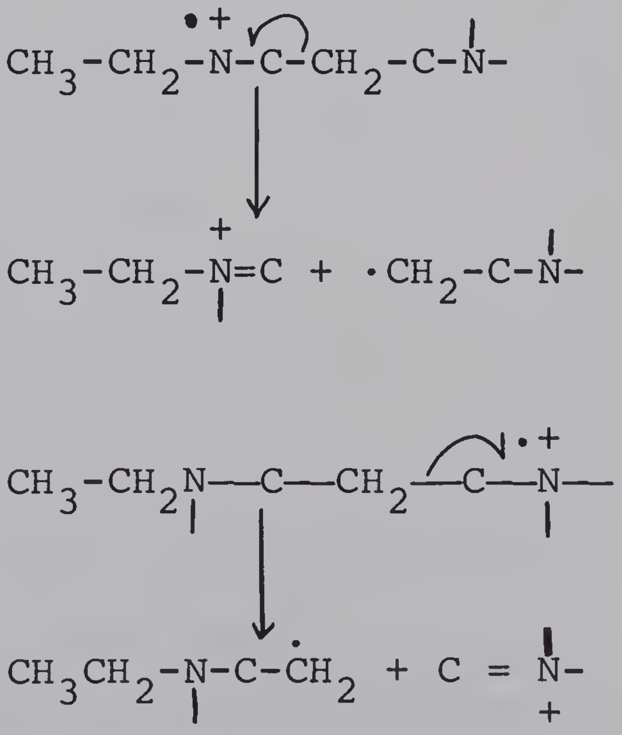

CH3
N
C=O
CH3
CH2
H
H
(c)
(c)
N
H

III


O
N-C-CH3
CH2
CH3
H
H
(c)
(c)
N
H

IV

The mass spectral fragmentation of tetrahydrodeoxylucidine-B into the ions at m/e 180 and 261 is then interpreted by a scheme analogous to the scheme shown below.



$$CH_3-CH_2-\overset{\bullet +}{N}-C-CH_2-C-N- \longrightarrow CH_3-CH_2-\overset{+}{N}=C + \cdot CH_2-C-N-$$

$$CH_3-CH_2N-C-CH_2-C-\overset{\cdot +}{N}- \longrightarrow CH_3CH_2-N-C-\dot{C}H_2 + C=\underset{+}{N}-$$

These results suggest that lucidine-A, lucidine-B and lycolucine possess structures which incorporate a 2,3,-6-trisubstituted pyridine skeleton as well as a 7-methylquinoline skeleton.

EXPERIMENTAL

Plant material was collected near Fredericton, New Brunswick, Canada, and identified by Mr. M. A. Stilwell.

Infrared spectra were recorded on a Perkin-Elmer Model 421 dual grating infrared spectrophotometer or a Perkin-Elmer Model 337 grating infrared spectrophotometer.

Nuclear magnetic resonance spectra were measured on Varian Associates Model A - 60 or Varian Associates Model HR-100 spectrometer with tetramethyl silane as an internal standard.

Optical rotatory dispersion spectra were measured on a Durrum-Jasco Recording Spectropolarimeter or Cary Model 60 Spectropolarimeter through the courtesy of Dr. C. Kaye. Optical activities were measured on a Perkin-Elmer Model 141 polarimeter.

Most of the mass spectra were determined on an A.E.I. Model MS-2H or an A.E.I. Model MS-9 mass spectrometer with heated inlet or direct probe (170-200°) and electron energy of 70 electron volts. Some mass spectra were measured on a Hitachi Perkin-Elmer RMU-6A mass spectrometer, through the courtesy of Dr. D. B. MacLean.

Gas-liquid chromatography was done on an Aerograph Model A-90-P3 gas chromatograph.

Alumina, unless otherwise specified, refers to basic alumina of activity II-III (Brockman scale). Research

Specialty Company aluminium oxide G was used for thin layer chromatography.

Dragendorrf's reagent was prepared according to procedure described by Bobbitt (93).

Microanalyses are by F. Pascher, Bonn, Germany; C. Daesle, Montreal, Canada; A. B. Gygli, Toronto, Canada or Mrs. D.Mahlow of this Department.

Ultraviolet spectra were recorded on Perkin-Elmer Model 202 Ultraviolet- Visible Spectrophotometer.

Melting points were determined on a Fischer-Johns melting point apparatus and are uncorrected.

THE ISOLATION OF THE TOTAL CRUDE ALKALOIDS

The total crude alkaloid mixture was obtained in these laboratories by Mr. J. McCutcheon using the procedure outlined below.

Finely ground plant material (5.1 kg) was percolated with methanol in a soxhlet extractor for three days. The solvent was evaporated off and the residue was digested with aqueous hydrochloric acid (6% by volume) and filtered. The residue was then digested with hydrochloric acid (10% by volume) and filtered. The filtrates were extracted with ether in order to remove non-basic material, basified to pH 10-11 with ammonium hydroxide and extracted with chloroform. The chloroform solution was dried over anhydrous sodium carbonate. Removal of the solvent under reduced pressure gave total crude alkaloids (55.6g).

SEPARATION OF ALKALOIDS INTO WEAK AND STRONG BASES.

(i) Dilute Aqueous Acetic Acid Method

The total crude alkaloid mixture (56.0 g) of L.lucidulum was dissolved in chloroform and extracted with aqueous acetic acid (5% by volume). After removal of the solvent from the chloroform layer a viscous oil (11.2g) was obtained. Residual acetic acid was removed by addition of an aqueous sodium bicarbonate solution to the residue and re-extraction with chloroform.

The weakly basic material (10.2 g) showed three major spots on t.l.c.with considerable tailing.The infrared spectrum in chloroform showed absorption at: 2790 (N-CH_3), 1700(very weak), and 1622 (amide, N-CO-) cm^{-1}. The n.m.r. spectrum was fairly well-defined, and showed a complex C-CH_3 signal in the τ 9.0 region and four signals in the τ 7.8-8.0 region due to N-CH_3 and N-CO-CH_3. The mass spectrum had peaks at m/e 205 (87; most intense peak above m/e 200), 218 (15), 233 (20), 247 (20), 280 (14), 261 (15), 273 (26), 455 (16), 465 (5), 481 (3), and 483 (3).

The pH of the aqueous layer was adjusted to 8 by addition of sodium bicarbonate. Extraction with chloroform gave more of the weakly basic material (39.9 g). Its t.l.c. behavior was identical with the 10.2 gram lot of weak bases already described.

The aqueous layer was finally basified with ammonium hydroxide. Extraction with chloroform gave strong bases (5.0 g).

(ii) Hydrochloric acid-Disodium Hydrogen Phosphate Method.

Total crude alkaloids (75.0 g) were dissolved in aqueous hydrochloric acid (10%). A solution of disodium hydrogen phosphate (0.2 M) was added from a buret until the pH of the solution was between 5.5 and 6.0 (pH paper). The aqueous solution was then extracted with methylene chloride (9 ℓ). On removal of solvent, a mixture of weak bases (31.5 g) was

obtained. The pH had then lowered to 4 and more of the phosphate solution was added until the pH was about 5.5. Extraction with methylene chloride (6 ℓ) gave more weak bases (7.9 g). The pH was adjusted to about 7 by further addition of disodium hydrogen phosphate. Extraction with methylene chloride and removal of the solvent afforded more weak bases (24.2 g).

The aqueous solution was then basified with dilute ammonium hydroxide and extracted with chloroform. Removal of solvent gave strong bases (18.8 g).

In later experiments, a saturated solution of disodium hydrogen phosphate (or the solid) was used in order to minmize the total volume and chloroform was used throughout instead of methylene chloride. The current procedure involves dissolving the crude alkaloids in 10-20% aqueous hydrochloric acid, adjustment of pH to 6-7 by addition of the phosphate solution, and several extractions with either methylene chloride or chloroform to give the weak bases. Basification of the aqueous solution with ammonium hydroxide and extracttion with chloroform or methylene chloride gives the strong bases.

The infrared spectrum of the weak bases had maxima at 2790 (N-CH_3), 1700 (shoulder), 1620 (amide; N-CO-), 1455, 1380 cm^{-1}. The n.m.r. spectrum had significant peaks at τ 7.75-7.90 (N-CH_3 and N-CO-CH_3) and τ 9.05 (C-CH_3- broad signal.)

The t.l.c. of the weak bases showed two major spots and several minor spots.

DRY-COLUMN CHROMATOGRAPHY OF WEAK BASES

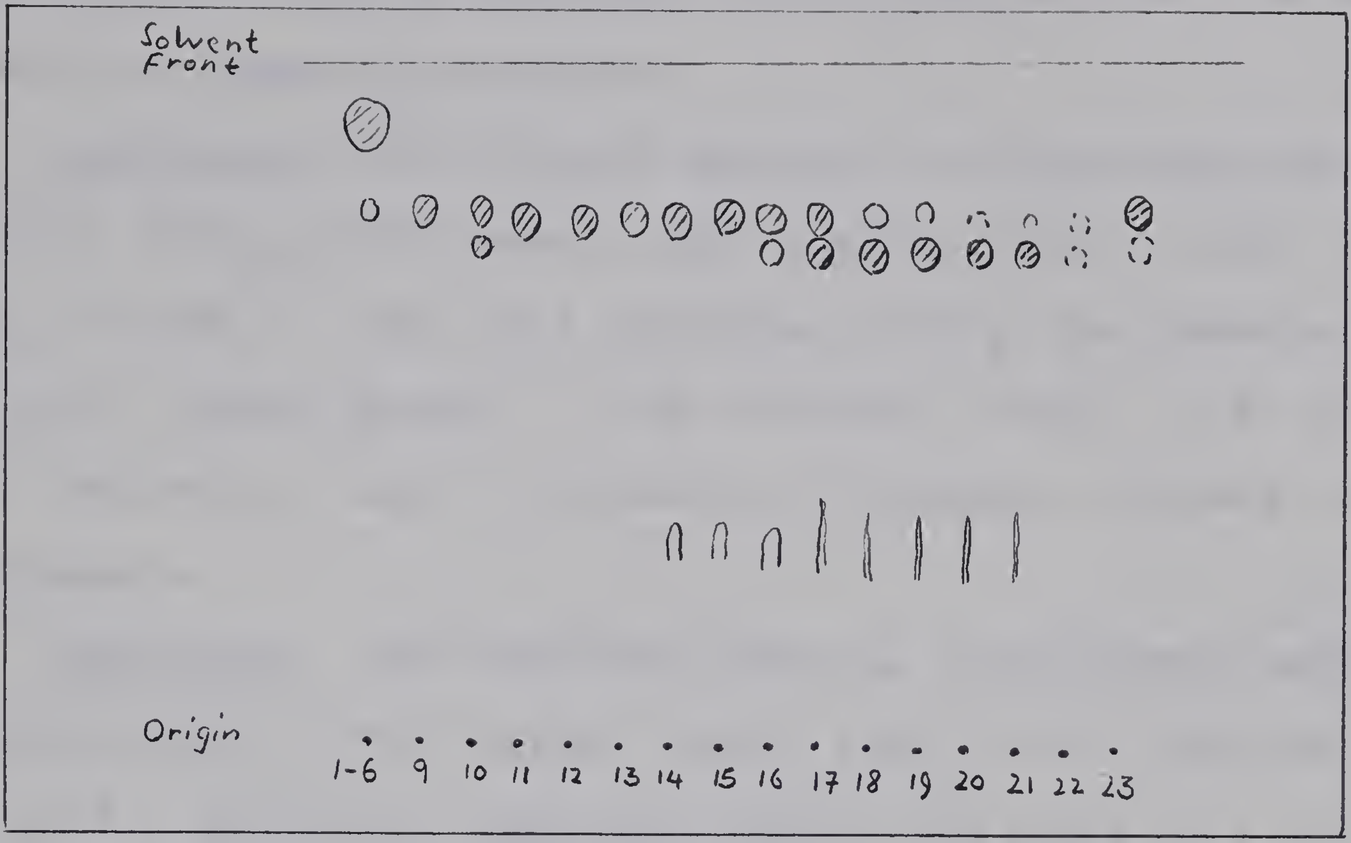
Polythene tubing with an internal diameter of 13 mm and about 60 cm long was fitted with a cork with a cotton-plugged glass tube outlet, and clamped vertically. The tube was filled with alumina (90 g -,BDH alumina, activity II-III). The weak bases (0.475 g) were added to the top of the alumina as a concentrated chloroform solution. A further layer of alumina (1 - 2 cm) was added followed by a 1-2 cm thick layer of sand.

The eluting solvent, ethyl acetate/ether (1:4), was added so that there was a constant head of 1-2 cm of liquid on top of the sand. Elution was complete when the solvent front reached the bottom of the alumina in the column. The polythene tubing was cut into 23 sections and the alumina extruded from each of the sections into appropriately numbered flasks. The alkaloidal content of each section was isolated from the alumina by elution with chloroform followed by ethyl acetate.

After work up, the fractions shown in the following table were obtained.

SECTION	Amount (mg)
1-6	10.0
7-8	0.0
9	5.0
10	13.5
11	6.6
12	5.1
13	48.0
14	2.1
15	11.0
16	36.6
17	59.7
18	49.8
19	24.0
20	11.1
21	2.2
22	6.2
23	21.5

The comparative t.l.c. of the above fractions on alumina with ethyl acetate-diethyl ether (1:1) as the eluting solvent is schematized in the following diagram.



Sections numbered 12, 15, 16, 19 and 20 were examined in more detail. Sections numbered 10-13 and 23 gave partly crystalline solids. The infrared spectrum of 12 was identical with that of lycopodine. T.l.c. also indicated that 23 contained lycopodine. The cause of the presence of lycopodine in sections as divergent in polarity as 12 and 23 has not been investigated.

Sections 15 and 16 contained mainly a single component with an amide band at 1620 cm^{-1} in its infrared spectrum (chloroform solution). We have named this compound lucidine-A.

Sections 19 and 20 contained mainly a more polar compound

with an amide band at 1620 cm^{-1} in its IR spectrum. We have named this compound lucidine-B.

Lucidine-A: The infrared spectrum in chloroform had bands at 2780 ($N-CH_3$) 1725 (weak), 1620 (amide, N-CO-), 1165, 1135, 1025, 960 cm^{-1}. The n.m.r. spectrum ($CDCl_3$) had peaks at τ 4.5-7.7 (minor peaks), τ 7.84 (singlet, $N-CH_3$), 7.92 and 7.95 ($N-CO-CH_3$), and τ 9.05 and 9.19 (complex six-peak $C-CH_3$ multiplet).

Lucidine-B: The infrared spectrum (chloroform) had bands at 2780 ($N-CH_3$) 1620 (amide, N-CO-) 1140, 1125, 1040 and 1025, 948 cm^{-1}. The n.m.r. spectrum ($CDCl_3$) had peaks at τ 4.5-7.5 - minor peaks; τ 7.83 ($N-CH_3$), τ 7.91 and 7.95 ($N-CO-CH_3$), and doublets at τ 8.93 and 9.16.

The t.l.c. of the more polar alkaloid, lucidine-B, indicated that it was more homogeneous than the less polar alkaloid, lucidine-A, and analysis was therefore carried out on a sample of lucidine-B prepared by evaporative distillation at 180-200° (0.5 mm).

Calculated for $C_{30}H_{49}N_3O$: C, 77.04; H, 10.56; N, 8.93.

Found: C, 75.76, 75.54; H, 11.66, 11.67; N, 7.33.

The mass spectrum of a sublimed sample of lucidine-B had the following peaks in the high mass range: m/e 467 (M,19.5%), 452 (M-15, 2.4%), 273 (47.1%), 260 (32.7%), 218 (10.7%), 217 (11%), 164 (12.4%) and 152 (9.6%). The base peak was at m/e 36.

COUNTER-CURRENT DISTRIBUTION OF WEAK BASES.

Preliminary equilibration experiments of the weak bases of L.lucidulum established the following partition coefficients between chloroform and the specified buffer solutions. The buffer solutions were prepared according to the procedures described by Vogel (50).

pH	Buffer Solution	$K \; \dfrac{\text{Amount in } CHCl_3}{\text{Amount in Buffer}}$
3.0	Potassium hydrogen phthalate and hydrochloric acid	2.9
3.0	Disodium hydrogen phosphate and citric acid.	1.1
3.0	Potassium hydrogen tartarate saturated solution.	1.9

The first counter-current distribution of the weak bases was done with the disodium hydrogen phosphate-citric acid (pH 3) buffer solution. In all later work, saturated potassium hydrogen tartarate was used as the buffer solution.

Only the first one hundred tubes of the Craig-Post 200 tube counter-current apparatus were used. Firstly, equal volumes (5000 ml) of each of the phases were shaken together for 3-5 minutes in lots in a separatory funnel (600 ml capacity) and separated. All of the moving phase (potassium bitartarate)

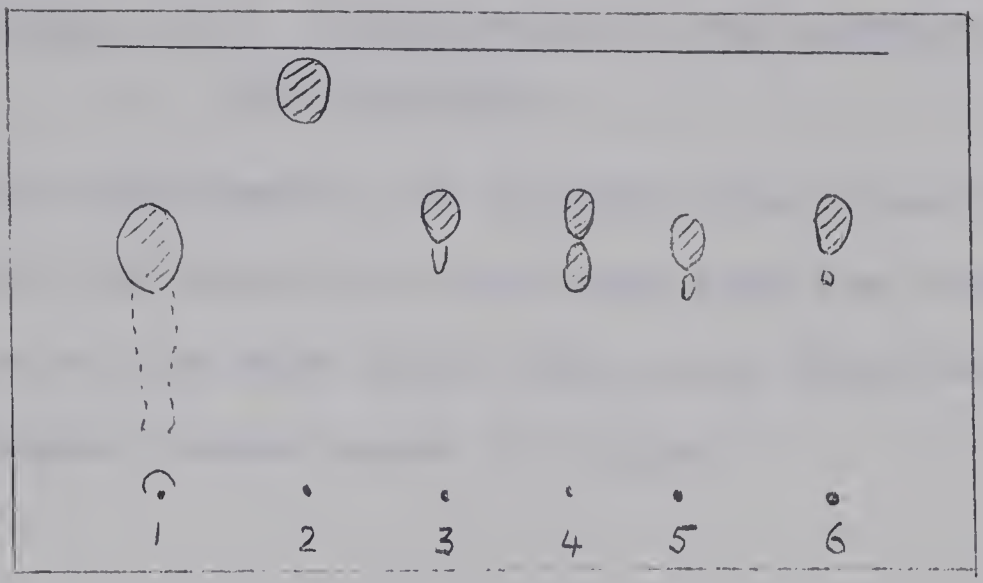
was added to the reservoir and 40 ml of the pre-equilibrated chloroform was added to each of the tubes 4-99. The robot was set to shake the tubes so as to mix the contents 20 times and to allow a two-minute period for the layers to separate. The sample of weak bases (5.1 g) was dissolved in pre-equilibrated chloroform (40 ml) and loaded into tube numbered 0. The rinsings from loading of tube 0 were put in tubes 1-3.

After the distribution was complete, t.l.c. analysis of the contents of tubes 0, 10, 20, 30, 40, 50, 60, 70, 80, 99 and also 64, 67, 71, 77, 81, 84, 87, 91, 94 and 97 was carried out. On the basis of the t.l.c.'s, the fractions tabulated below were collected.

AMOUNT WEAK BASES: 5.1 g

Fraction	1	2	3	4	5	6
Tubes	0-4	5-44	45-52	53-66	67-93	94-99
Amount(g)	0.621	0.258	0.263	1.708	0.948	0.205
Some IR Maxima (cm^{-1}) ($CHCl_3$)	2780 1700 1620	2780 1700 1620	2783 1620	2783 1620	2783 1620	1691 1620

The comparative thin layer chromatography of the above fractions (on alumina with ethyl acetate-diethyl ether (1:1) as the eluting solvent) is shown in the following figure.



Infrared spectra and t.l.c. behavior of fractions 1-6 led us to make the following inferences:

Fractions 3 and 5 were rich in lucidine-A and lucidine-B respectively, while fraction 6 was mainly lycopodine. Fraction 1 contained alkaloids with a band at 1620 cm^{-1} in the infrared spectrum. We have tentatively named this mixture 1620 H. Fraction 2 contained an alkaloid which is less polar than lucidine-A, lucidine-B and 1620 H.

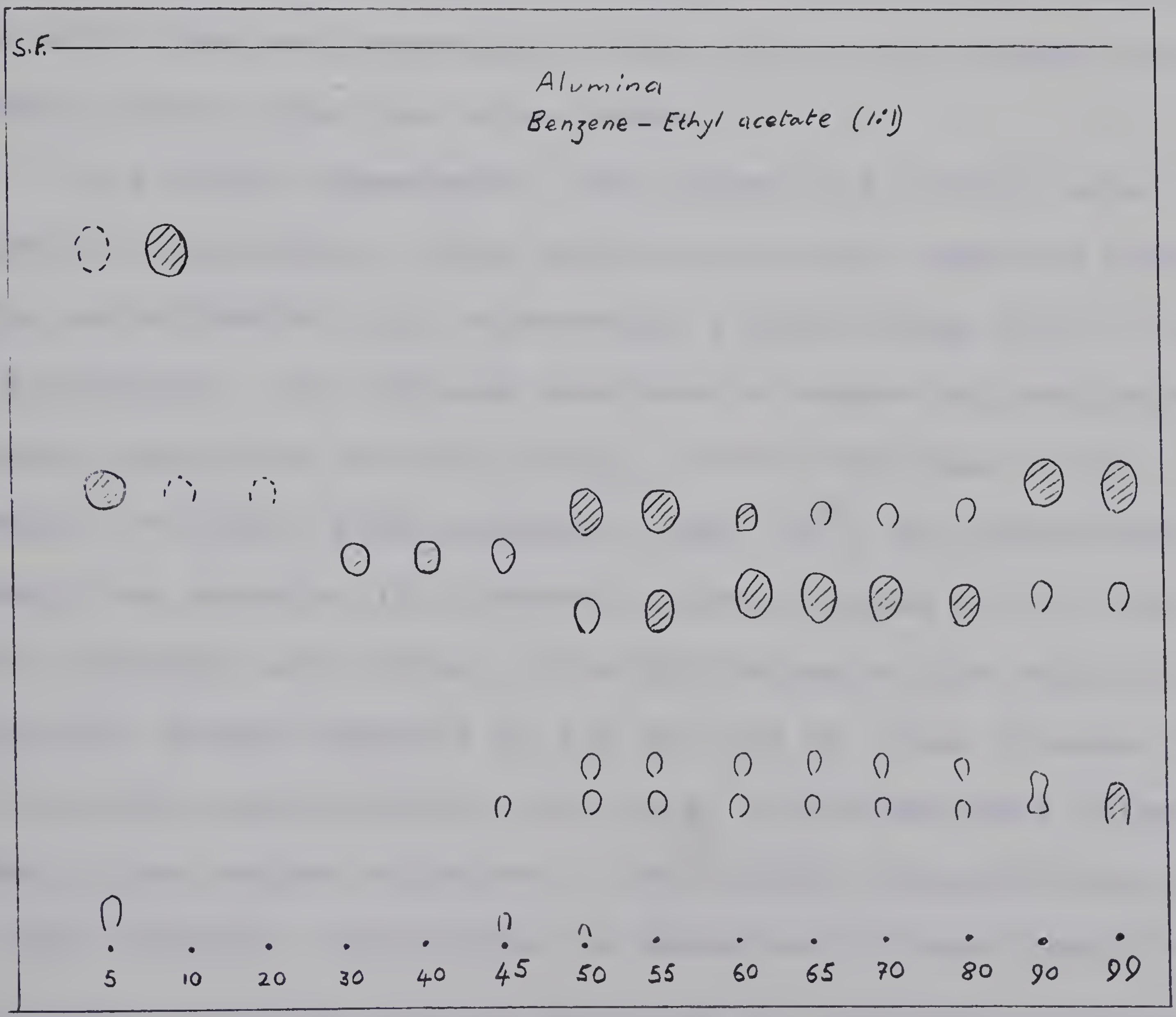
Dry-column chromatography of Fraction 2 gave a slightly yellowish oil (66.3 mg) and some compounds (18.7 mg) with an amide band at 1620 cm^{-1} in the i.r. spectrum.

The infrared spectrum (chloroform) of the oil, had bands at 2793 (N-CH_3), 1693 ($>$C=O), 1468, 1459, 1400 (-CH_2-CO-), cm^{-1} and a well-resolved fingerprint region. We originally referred to this alkaloid as "1690 Ketone", because of the strong absorption at about 1690 cm^{-1} and have recently named

it luciduline.

A MORE DETAILED T.L.C. EXAMINATION OF THE COUNTER-CURRENT DISTRIBUTION.

In later experiments, we observed that there were more components in the fractions collected from the counter-current distribution of the weak bases than we at first believed. The chromatogram shown below is typical.


S.F.
Alumina
Benzene – Ethyl acetate (1:1)
5
10
20
30
40
45
50
55
60
65
70
80
90
99

Tube 5 has three spots, two of which correspond to luciduline (R_f = 0.7) and 1620 H (R_f = 0.5).

Tube 10 contains mainly luciduline. The single component of tubes 30, 40 and 45 has been shown to be a new alkaloid and we have named it lycolucine. Tubes 50-80 contain not only lucidine-A and lucidine-B in varying ratios, but also two hitherto undetected compounds which have the 1620 cm^{-1} amide band and which we have tentatively named 1620 C and 1620 D. Further t.l.c. studies indicate that the 1620 C and D portion has more than two components. Tubes 90 and 99 contain lycopodine and at least two other bases.

In a recent experiment, the contents of tube 30 were examined separately. After basification with ammonium hydroxide and extraction into chloroform, a white foamy solid (12 mg) was obtained. Its infrared spectrum in carbon tetrachloride showed absorption at 2775 ($N-CH_3$), 1702 (very weak), 1651 (amide, $N-CO-CH_3$) 1580 (aromatic ring) cm^{-1}. Its ultraviolet absorption spectrum (95% ethanol) showed maxima at 217, 261, 272 (shoulder) and 300 mμ. On acidification of the ethanolic solution, maxima appeared at 258 and 268 mμ (less intense than 261 and 272 respectively), and 320 mμ (broad and more intense than in the neutral solution). The further characterization of this alkaloid, lycolucine, is described in more detail below.

A GENERAL PROCEDURE FOR THE ISOLATION OF LUCIDINE-A, LUCIDINE-B and 1620 C/D.

The t.l.c. behavior of the middle tubes in the counter-

current distribution was carefully examined in order to exclude the use of lycolucine-containing fractions in the dry-column chromatographic separation of lucidine-A, lucidine-B and 1620 C/D. If necessary, the u.v. spectrum of the sample from an individual tube was taken in order to see if there was any lycolucine present. A careful check was also made to exclude the use of samples that might contain even small amounts of lycopodine.

The weak bases, free from both lycolucine and lycopodine, were then subjected to dry column chromatography with 1:1 benzene/ethylacetate or 1:1 ether/ethyl acetate as solvent on 200-340 g of alumina (BDH or Fisher basic alumina activity II/III) in polythene tubing (internal diameter 20 mm). The contents of the 2-3 cm cut-and-then-extruded sections were compared by t.l.c. and six or more fractions with the specified compositions were collected as illustrated in the following summarized experiment. The R_f values are based on the distance of the section from the solvent front and are used merely to indicate the relative positions of the various fractions.

Rechromatography (dry-column) of the lucidine-A-rich fraction 1 gave pure (by t.l.c.) lucidine-A. Lucidine-B was obtained in a similar way. Except for the preliminary examination reported later in this section, 1620 C/D mixtures were reserved for future investigation.

Fraction	R_f value	Composition
1	0.39 - 0.45	Mainly lucidine-A
2	0.36	Mainly lucidine-A; some lucidine-B.
3	0.33	Lucidine-A and lucidine-B in equal amounts.
4	0.27 - 0.30	Mainly lucidine-B; some lucidine-A.
5	0.23	Mainly lucidine-B.
6	0.03 - 0.18	1620 C and 1620 D

Lucidine-A so obtained was a white solid (foam) with optical rotation $[\alpha]_D$ - 21° (c,0.20, chloroform). Ultraviolet spectrum (95% ethanol: strong end absorption and weak bands at 265 mμ(broad) and 305 mμ.

Mass spectrum (Fig.15): prominent peaks at m/e 467 (molecular ion, 17), 273 (100), 260 (14), 218 (8), 217 (10), 166 (8), 184 (12), 163 (13). Infrared spectrum (Fig.8): $\nu_{max}^{CHCl_3}$, 2790 (N-CH_3), 1620 (N-CO-CH_3), cm^{-1} and a weak shoulder at 1700 cm^{-1}. Nuclear magnetic resonance spectrum (Fig. 5): τ 5.0-7.8 (minor peaks), 7.86 and 7.87 (two peaks, N-CH_3), 7.94 and 7.97 (N-CO-CH_3), 9.15 (complex C-methyl multiplet).

In perdeuteroacetic acid, the N-CH_3 signal is shifted to lower field (τ 7.20).

Lucidine-B, is a white solid (foam), with optical rotation, $[\alpha]_D$ - 46° (C, 0.23, chloroform). Ultraviolet spectrum (95% ethanol): shows only strong end absorption. Optical rotatory dispersion (sample purified by evaporative distillation) in methanol (C, 0.08): $[\alpha]_{260}$ - 138,000 $[\alpha]_{230}$ + 320,000.

Its infrared spectrum (Fig. 9): $\nu_{max}^{CHCl_3}$, 2790(N-CH_3), 1621 (N-CO-CH_3), 1460, 1380 (doublet, CH_3-C) and characteristic maxima at 1138, 1125, 1102, 1040 and 1027 cm^{-1}. Nuclear magnetic resonance spectrum ($CDCl_3$): τ 4.5 - 7.50(minor peaks; 7.83 (N-methyl),7.91 and 7.95 (N-CO-CH_3) and doublets at τ 8.93 and 9.16 (Fig.6). In perdeuteroacetic acid, the N-methyl peak is at τ 7.20. The mass spectrum of lucidine-B (Fig.16) shows prominent peaks at m/e 467 (44, molecular ion), 452 (5, M-15), 273 (100), 260 (81), 218 (23), 217 (26), 164 (17), and is similar to that of the alkaloid lucidine-A.

The exact mass of the molecular ion is 467.3870 and corresponds to $C_{30}H_{49}N_3O$ (Calculated: 467.3875).

1620 C/D Mixture

The 1620 C/D mixture obtained was a white foamy solid. Infrared spectrum: 3350 (broad,-OH); 2898 (N-CH_3), 1640 (amide $\rangle$C=O), 1450, 1420, 1380, 1370, 1260. Nuclear magnetic resonance spectrum ($CDCl_3$): τ4.8-7.5 (minor peaks) 7.75, 7.90, and 7.93 (N-CH_3 and N-CO-CH_3) 8.70, 9.10 (CH_3-C, multiplet). Mass spectrum: peaks at m/e 483 (70), 465 (70), 455 (77),

289 (27), 287 (27), 276 (50), 271 (84), 258 (32), 233 (66). A metastable at m/e 448.0 corresponds to m/e 483 losing 18 mass units forming m/e 465.

The following exact masses were determined:

Composition	Mass Calculated	Mass Observed
$C_{30}H_{49}N_3O_2$	483.3825	483.3809
$C_{30}H_{47}N_3O$	465.3719	465.3704
$C_{29}H_{49}N_3O_2$	455.3875	455.3889
$C_{17}H_{23}N_2O_2$	271.1810	271.1797
$C_{18}H_{27}N_2$	271.2174	271.2167

Analysis: Calculated for $C_{30}H_{49}N_3O_2$: C, 74.49; H, 10.21; N, 8.69. Found: C, 73.40; H, 10.59; N, 8.74.

GENERAL PROCEDURE FOR THE ISOLATION OF LYCOLUCINE

The counter-current distribution fractions which contain lycolucine (t.l.c. and u.v.), are combined and chromatographed by the dry-column method. After careful t.l.c. examination of the contents of the section, pure lycolucine (one spot on t.l.c.) was obtained. In one experiment, 9.7 grams of weak bases gave 0.364 g of crude lycolucine which on dry-column chromatography gave pure lycolucine (140 mg).

Lycolucine is a white solid, optical rotation, $[\alpha]_D$ + 399° (C, 0.73 chloroform). Infrared spectrum (Fig.11): 2780 ($N-CH_3$), 1640, ($\nu_{max}^{CCl_4}$), 1580, 1560, 1460, 1450, 1435, 1420, 1375, 1370 cm^{-1}. Ultraviolet spectrum: $\lambda_{max}^{95\%\ ethanol}$, 217 ($\epsilon$ = 14,530), 261 (ϵ = 8,800), 272 shoulder (ϵ=7,824) and 300 (ϵ = 10,200).

On acidification, 210 mμ (less intense than 217 mμ) 258 mμ and 268 mμ (less intense than 261 and 272 mμ respectively), 320 mμ (more intense and broader than 300 mμ). Nuclear magnetic resonance ($CDCl_3$): τ 2.70 and 3.20 (2H quartet J=8 cps), 3.43 (1H Ar-CH=C or Ar-C=CH-) 7.88 and 7.93 ($N-CH_3$ and $N-CO-CH_3$), 9.10 (CH_3-C, 6H).

Mass spectrum (Fig. 17): m/e 461(molecular ion, 69), 446 (M-15, 17) 268 (100), 185 (20), 164 (12), 164 (12) and 150 (27).

A sample of lycolucine which was isolated by the above method has recently been crystallized from acetone solution by Mr. L. F. Ball, in these laboratories. The crystalline lycolucine isolated in this way has m.p. 198°-200°C and shows spectral properties very similar to those reported above.

ISOLATION OF LUCIDULINE

The isolation of luciduline from counter-current distribution fractions by dry-column chromatography has been described above (page 99). The final step in the purification is accomplished by evaporative distillation at 80° - 100° under vacuum

(0.5 mm). Luciduline is a clear colorless oil which becomes yellow on standing. It forms a crystalline, hygroscopic perchlorate m.p. 194-196°C when 60% perchloric acid is added to a methanolic solution of the base. Crystallization occurs after a small amount of ether is added and the solution is kept in the fridge for 1-2 days.

Infrared spectrum (Fig.11): $\nu_{max}^{CHCl_3}$, 2780 ($N-CH_3$), 1690 ($>C=O$), 1400 (CH_2-CO-) cm^{-1}. Optical rotatory dispersion in ethanol (C, 0.283): $[\alpha]_{308}$ + 2403, $[\alpha]_{268}$ -2863. Ultraviolet spectrum (95% ethanol): strong end absorption with shoulder at 285 mμ. Nuclear magnetic resonance spectrum ($CDCl_3$, Fig.1): τ 6.94, 7.22, 7.62, 7.78, 7.87 ($N-CH_3$), 9.12 (CH_3CH, doublet J=6 cps). Mass spectrum (Fig.12): m/e 207(80), 206(42), 192 (19), 164(100), 150(27), 96(87), 70(34), 55(51), 44(65), 42 (70) and 41 (44). The molecular ion corresponds to $C_{13}H_{21}NO$ (Calculated for $C_{13}H_{21}NO$: 207.1630. Found: 207.1630).

Analysis: Calculated for $C_{13}H_{21}NO$: C, 75.31; H, 10.21. Found: C, 74.03; H, 10.03.

DIHYDROLUCIDULINE

Luciduline (21 mg) was dissolved in methanol (15 ml) and cooled in an ice-bath. Excess sodium borohydride (300 mg) was added to the ice-cold solution and stirred magnetically for four hours. T.l.c. analysis of an aliquot of the reaction mixture taken 3-4 minutes after addition of sodium borohydride showed that the reaction was complete. Addition of water (ice-cold) and extraction with chloroform gave dihydroluciduline (13 mg) as a white crystalline solid. Dihydroluciduline was purified further by evaporative distillation under vacuum at 80 - 110°C. Analysis. Calculated for $C_{13}H_{23}NO$:C, 74.59; H, 11.07; N, 6.69. Found: C, 74.53; H, 10.93; N, 7.07. PK^{*}_{mcs}, 7.7. Infrared spectrum: $\nu_{max}^{CHCl_3}$, 3605 (-OH), 2780 ($N-CH_3$) 1460, 1380, 1370, 1355 cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$ (100 Mcs): τ 6.05 (multiplet, $W_{\frac{1}{2}h}$ = 23 cps. IH), 6.75, 6.88, 7.48, 7.60, 7.75 (multiplets, 3H) 7.86 (singlet, $N-CH_3$), 9.14 (doublet, splitting 7 cps, CH_3CH). Shaking with D_2O did not reveal the chemical shift of the -OH proton. Nuclear magnetic resonance spectrum in perdeuteroacetic acid (100 Mcs): τ 5.98 (multiplet), 6.65 and 6.75, 6.94, 7.05 ($N-CH_3$, singlet). Irradiation experiments show that the τ 6.94 signals are coupled to τ 5.98; τ 9.08 (CH_3 doublet, splitting 6 cps; coupled to τ 8.10 region).

Mass spectrum: m/e 209 (molecular ion, 26), 208 (43), 192 (Base Peak), 166 (29), 164 (18), 96 (11), 70 (12) and 44 (33). A metastable peak at m/e 177.5 corresponds to the fragmentation m/e 209 → m/e 192.

High resolution mass spectral data.

Molecular formula.	Calculated.	Found.
$C_{13}H_{23}NO$	209.1779	209.1771
$C_{13}H_{22}N$	192.1752	192.1749
$C_{11}H_{18}N$	164.1439	164.1439
$C_{10}H_{16}NO$	166.1232	166.1237

Infrared - H-bonding studies on the alcohol: the -OH absorption at 3603 cm^{-1} remains unchanged on dilution.

O-ACETYLDIHYDROLUCIDULINE

Dihydroluciduline (54 mg) was dissolved in a mixture of acetic anhydride (1 ml) and pyridine (0.5 ml) and the mixture was allowed to stand at room temperature for 12 hours. After work-up, O-acetyldihydroluciduline (63 mg) was obtained as a colorless oil which was purified by molecular distillation under vacuum. O-acetyldihydroluciduline is less polar than both luciduline and dihydroluciduline (t.l.c). Infrared spectrum: $\nu_{max}^{CCl_4}$, 2779 ($N-CH_3$), 1733 (ester

carbonyl) 1455, 1443, 1378, 1370, 1235, 1130, 1026 $cm.^{-1}$ Nuclear magnetic resonance spectrum in $CDCl_3$: τ 5.03 (multiplet, $W_{\frac{1}{2}h}$ = 24 cps, IH); 6.93 (IH, doublet, splitting 11 cps), 7.44 (doublet of doublet, splittings large, about 12 cps), 7.92, 7.98 (singlets, $N-CH_3$ and CH_3-CO-O); 9.17 (doublet, splitting 7 cps).

Irradiation experiments show that the τ 5.03 proton is coupled to the τ 8.33 region and also to τ7.44 (splitting ca 12 cps). The τ 6.93 proton is coupled to the τ 7.80 region (11 cps coupling). The secondary methyl is coupled to a proton in the τ 8.03 region.

Nuclear magnetic resonance in perdeuteroacetic acid: τ 5.00 (multiplet, $W_{\frac{1}{2}h}$ = 23 cps, IH); 6.08 (doublet, J = 13.5 cps; IH, coupled to the proton at τ 6.80) 6.60 (broad singlet, IH), 6.80 (doublet split into doublet, J = 13.5 and 3.5 cps; IH). The τ 6.80 proton is coupled to τ 6.08 (J = 13.5 cps) and τ 7.87 (J = 3.5 cps). τ 7.00 (singlet, NCH_3), 7.94 (singlet, CH_3-CO-O) 9.04 (doublet, J = 6 cps, CH_3CH). Mass spectrum: m/e 251 (5), 250 (4), 208 (4), 192 (100), 150 (4). The molecular ion has an exact mass of 251.1897 which corresponds to the molecular formula $C_{15}H_{25}NO_2$ (calculated, 251.1885).

DEUTERIUM EXCHANGE WITH LUCIDULINE

Freshly distilled luciduline (66 mg) was dissolved in a solution of deuterium chloride in acetic acid-O-d (5 ml), and allowed to stand at room temperature for five days. The bulk of the solvent was removed under reduced pressure and the residue was basified by addition of a saturated heavy water solution of anhydrous potassium carbonate (6 ml). Extraction with deuterochloroform (9 ml), followed by drying the extract over anhydrous sodium sulfate and the removal of the solvent gave luciduline-d_2 (40 mg).

Infrared spectrum: $\nu_{max,}^{CHCl_3}$ 2785 ($N-CH_3$), 1690 (C=O), 1460, 1452 cm.$^{-1}$. The band at 1400 cm.$^{-1}$ present in luciduline was no longer apparent. Nuclear magnetic resonance spectrum in $CDCl_3$: No signals below τ 7.00. τ 7.11 and 7.23 (IH, doublet split into doublet, J = 12 cps and 4 cps). The rest of the N.M.R. spectrum is similar to that of luciduline. Its N.M.R. spectrum in perdeuteroacetic acid is similar to that of the undeuterated luciduline.

DIHYDROLUCIDULINE-D_2

Luciduline-d_2 (40 mg) was dissolved in methanol-O-d (2 ml) and excess sodium borohydride (about 50 mg) was added to the cooled and well-stirred solution. The

mixture was stirred for a further two hours. After the usual work-up, dihydroluciduline-d_2 was obtained. Mass spectrum: m/e, 211(27),210 (44), 195 (19), 194 (100), 193 (29) 168 (26), 164 (18), 150 (12), 98 (10), 70 (10), 44 (18).

O-ACETYLDIHYDROLUCIDULINE-D_2

The dihydroluciduline-d_2 obtained by the sodium borohydride reduction described above, was dissolved in acetic anhydride (2ml) and pyridine (1 ml) and allowed to stand at room temperature for 21 hours. After the usual work-up, O-acetyldihydroluciduline-d_2(35 mg) was obtained as an oil. A sample obtained by evaporative distillation was used to obtain spectral data. Infrared spectrum: $\nu_{max}^{CCl_4}$ 2779 (N-CH_3), 1732 (ester carbonyl), 1455, 1370, 1235, 1028 cm.$^{-1}$ Mass spectrum: m/e 253 (5), 252 (5), 195 (20), 194 (100) and 193 (20). The large M-1 peak associated with the molecular ion made the peaks associated with the molecular ion unsuitable for the calculation of the percentage of deuteration (73). The calculation of the percentage of mono-, di-, tri, tetra- and undeuterated species was therefore done on the basis of the M-59 peak at m/e 194. The calculation indicated the absence of undeuterated and tetradeuterated species and the presence of 84% of d_2, 14% of d_1 and 6% of

d_3 species. Nuclear magnetic resonance spectrum in $CDCl_3$: Similar to the undeuterated O-acetyldihydroluciduline except for the absence of the τ 7.44 signal and the reduced multiplicity of the τ 5.0 signal; τ 5.04 (broad, IH; sharpened by irradiation at τ 8.09 and τ 7.81), 6.96 (doublet, J = 11 cps; IH; coupled to τ 7.81), 7.90 and 7.97 (two singlets $N\text{-}CH_3$ and O-Acetyl), 9.17 (methyl doublet, J = 6.5 cps; coupled to τ 7.81 region).

Nuclear magnetic resonance spectrum in perdeuteroacetic acid: τ 4.98 (doublet, J = 4.5 cps, IH; coupled to τ 7.74 region); the rest of the spectrum from τ 5.0 - 9.5 is similar to that of undeuterated O-acetyldihydroluciduline. Simultaneous irradiation of the τ 7.74 region collapses the τ 4.98 doublet to a singlet and the τ 6.81 quartet to a doublet (J = 13 cps).

DIHYDROLUCIDULINE-D_1

Luciduline (5 mg) was dissolved in methanol (5 ml) and reduced with sodium borodeuteride (19 mg) in the usual way. After work-up, the dihydroluciduline-d_1 (about 3 mg) obtained was sublimed. Mass spectrum: m/e 210 (30), 209 (45), 194 (16), 193 (100), 192 (18), 167 (27), 164 (19), 150 (6), 96 (11), 70 (10), 57 (11), 44 (56).

SELENIUM DEHYDROGENATION OF DIHYDROLUCIDULINE

Dihydroluciduline (25 mg) was mixed with selenium powder (200 mg) and heated in a sealed tube (sealed under vacuum) at 280-299° for 8 hours. The cooled dehydrogenation mixture was isolated by direct sublimation from the dehydrogenation tube. The product obtained after sublimation under vacuum at 90° for 3 hours was a mixture of solid and liquid material. Ultraviolet spectrum: $\lambda_{\max}^{\mathrm{EtOH}}$, 227 (intense and sharp), 276, 309, 311 and 324 mμ. Acidification of the ethanolic solution caused practically no change in the u.v. spectrum. Mass spectrum: m/e 170 (94), 156 (100), 155 (71), 141 (40). Gas-liquid chromatographic analysis showed the presence two major components approximately in the ratio 3 to 1 and four very minor components. Attempts to collect the minor components were unsuccessful, but the u.v. spectrum of the combined minor components was similar to that of the mixture.

The two major components were isolated by gas-liquid chromatography on 10% QF-1 on Chrom W (60-80 mesh) in a 5' x 1/4" column at a temperature of 125° and a helium flow rate of 35 ml/min. The major component, retention time 8.7 min., was isolated as a white crystalline solid. Mass spectrum: m/e 156 (100), 155 (31), 141 (55) and a metastable peak at m/e 127.5 corresponds to the

fragmentation of m/e 156 to m/e 141. The exact mass of the molecular ion corresponds to the molecular formula $C_{12}H_{12}$ (Calculated, 156.0943. Found, 156.0940). The m/e 141 peak analyses for $C_{11}H_9$ (Calculated, 141.0706. Found, 141.0704). Ultraviolet spectrum: $\lambda_{\max}^{\mathrm{EtOH}}$, 275 and 311 mμ.

The above ultraviolet spectrum was compared with the ultraviolet spectra of 1,5-, 1,8-, 2,3-, 2,6-, and 2,7-dimethylnaphthalenes. The ultraviolet spectrum of the dehydrogenation product was very similar to that of 2,6-dimethylnaphthalene. The mass spectrum of 2,6-dimethylnaphthalene is practically identical with that of the dehydrogenation product.

The minor component was obtained as an oil and had a retention time of 14.2 minutes under the conditions described above. Ultraviolet spectrum: $\lambda_{\max}^{\mathrm{EtOH}}$, 229, 280, 311, 317, 326 mμ. Mass spectrum: m/e 170 (100), 169 (19), 155 (52). Exact mass determination shows that the m/e 170 and 155 have the molecular compositions, $C_{13}H_{14}$ (Calculated, 170.1096. Found, 170.1092.) and $C_{12}H_{11}$ (Calculated, 155.0860. Found, 155.0856).

SELENIUM DEHYDROGENATION OF LUCIDULINE

Freshly distilled luciduline (30 mg) was mixed with

selenium (100 mg) in a thick-walled Carius tube, sealed under vacuum and heated at 300-310°C for 16 hours. Direct sublimation of the dehydrogenation mixture at 116 - 120° under vacuum (5 mm) gave a solid sublimate (9 mg) in 43% yield. Ultraviolet spectrum: $\lambda_{\text{max.}}^{\text{EtOH}}$, 227, 274, 308, 324 mμ. There is no change on basification with dilute aqueous sodium hydroxide, and only very slight change occurs on acidification with hydrochloric acid. Mass spectrum: m/e 170, 156, 155, 141, 128, 115, 77 and 76; it is similar to the mass spectrum of the dehydrogenation products of dihydroluciduline. Gas-liquid chromatographic analysis revealed the presence of three components, called A,B,C in the ratio 4:91:5. Using a 5' x 1/4" SE 30 (20%)on Chromosorb W 60-80 column at 168° and a flow rate of 24 ml/min. A,B, and C had the retention times of 11.0, 15.5 and 23.0 mins. respectively.

The major component, B, isolated by gas-liquid chromatography is a solid. Infrared spectrum: $\nu_{\text{max}}^{\text{CCl}_4}$, 3020, 3010, 2918, 1910, 1753, 1609, 1500, 867 cm^{-1}. The infrared spectrum is practically identical with that of 2,6-dimethylnaphthalene. Mass spectrum: m/e 156 (100), 155 (31), 141 (42), 77 (12), metastable peak at m/e 127.5.

The minor component, C, has a mass spectrum with peaks at m/e 170 (100), 168 (16), 155 (54) and a metastable peak at 141.5 corresponding to m/e 170 → m/e 155. Ultraviolet

spectrum, $\lambda_{\text{max.}}^{\text{EtOH}}$: 225, 278, 288, 311 325 mμ. The quantity of the minor component A was not sufficient for an ultraviolet spectrum.

ATTEMPTED PHOTOCHEMICAL CLEAVAGE OF LUCIDULINE.

(i) In methanol.

A solution of luciduline (38 mg) in methanol (3 ml) was flushed with nitrogen for several minutes and then irradiated with a Hanovia high pressure mercury lamp (57 A 36) for three days. Gas chromatography of the reaction mixture on a 10' x 1/4" column packed with QF-1 10% on Chromosorb W (60-80 mesh) at 200°, with a helium flow rate of 60 ml/min showed the presence of four components as summarized below:

Peak	1	2	3	4
Retention Time (Min.)	7.3	10.4	14.6	23.0
Relative Area	1	1	10	1
Compound	Unknown	Unknown	Luciduline	Unknown

(ii) In hexane.

A solution of luciduline (30 mg) in hexane (3 ml) was irradiated for 3 days with a Hanovia high pressure mercury lamp (57A36.) A flocculent white precipitate which formed

was centrifuged off and washed with hexane. The residue (1 mg) failed to give a mass spectrum. Gas chromatography of the rest of the reaction mixture indicated the formation of a mixture of products similar to the one obtained in the photolysis of luciduline in methanol.

ATTEMPTED BAEYER-VILLIGER OXIDATION OF LUCIDULINE

The method used was a modification of the one described by Meinwald and co-workers (74). m-Chloroperbenzoic acid (20 mg) was added to a solution of luciduline (13.7 mg) in dry benzene (5 ml). The flask containing the mixture was covered with aluminum foil and placed in a dark cupboard for five days. After removal of benzene under reduced pressure, aqueous sodium sulfite (about 2 ml) and then sodium bicarbonate (about 2 ml.) were added . The mixture was extracted with benzene which was then dried over anhydrous sodium carbonate. Removal of the solvent gave a residue (5 mg). The residue was shown to be unchanged luciduline (t.l.c. and infrared spectrum).

When the oxidation was repeated in dry methylene chloride instead of benzene similar results were obtained.

BROMINATION OF LUCIDULINE

Luciduline (309 mg,) was dissolved in a saturated solution of hydrogen bromide in methanol (2 ml). After 2-3 hours, the methanol was removed under reduced pressure,

and the residue was dissolved in chloroform (2 ml). A solution of bromine in chloroform (385 mg in 2.2 ml.) was then added, dropwise. The initially formed yellowish precipitate dissolved when the color of bromine disappeared. At the end of the reaction there were two immiscible layers in the flask. The reaction mixture was stirred for a further two hours. After the removal of the solvent under reduced pressure, excess sodium bicarbonate was added to the residue until no effervescence occurred on further addition of the bicarbonate solution. Extraction with chloroform, followed by the usual work-up gave a white crystalline solid (119 mg). Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2770 ($N-CH_3$), 1670 ($>C=O$, conjugated), 1603 (strong, $C = C$), 1450 cm^{-1}. Ultraviolet spectrum: λ_{max}^{EtOH}, 213 mμ ($\epsilon = 5346$) 263 mμ ($\epsilon = 7350$). Mass spectrum: 285 (59), 284 (15), 283 (61), 243 (98), 242 (16), 241 (100), 205 (17), 204 (26), 162 (44); and 98.5 (metastable peak).

The exact mass of the molecular ion corresponds to the molecular formula, $C_{13}H_{18}NOBr$ (Calculated, 285.0549. Found, 285.0549). The m/e 243 peak analyses for $C_{10}H_{12}NOBr$ (Calculated, 243.0083. Found, 243.0084).

Nuclear magnetic resonance spectrum in $CDCl_3$: τ 6.84 (doublet, J = 7 cps, IH), 7.10. 7.20, 7.32, 7.46 (multiplets, 3H), 7.88 (singlet, $N-CH_3$), 8.95 (CH_3 doublet, J = 7 cps). Irradiation at τ 8.01 collapses the doublet at τ 6.84 to a

singlet and the methyl doublet at τ 8.95 to a singlet. Nuclear magnetic resonance in perdeuteroacetic acid: a poorly resolved spectrum was obtained due to the low solubility of the compound in perdeuteroacetic acid; τ 6.12, 6.33, 6.40, 6.97 (singlet, N-CH_3), 8.82 (CH_3 doublet, J = 5 cps).

DEHYDROLUCIDULINE

Freshly distilled lucidulíne (136 mg, 0.66 m mole) was refluxed with freshly sublimed selenium dioxide (100 mg, 0.9 m mole) in dioxane (5 ml) for 22 hours. The mixture turned dark brown within a few minutes. The cooled reaction mixture was filtered and the residue washed with chloroform. After removal of solvents under reduced pressure, the brownish residue was redissolved in chloroform and the chloroform solution was extracted twice with dilute aqueous sodium bicarbonate. Work-up afforded dehydroluciduline (64 mg). Further purification was effected by molecular distillation under vacuum at 150°. The distillate solidified on cooling. Infrared spectrum: $\nu_{max}^{CCl_4}$, 2780 (N-CH_3), 1668 (C = O), 1628 (C = C), 860 cm^{-1}.

Nuclear magnetic resonance spectrum in $CDCl_3$: τ 3.95 (sharp singlet, IH, vinyl proton), 7.12 (doublet, J = 11 cps, further split into triplets, J = 2.5 cps, IH), 7.86 (singlet, N-methyl), 8.30 (doublet, J = 11 cps, further split into triplets, J = 2.5 cps, IH), 8.97 (doublet, J = 6 cps, C-methyl).

Irradiation at τ 8.00 causes the methyl doublet at τ 8.97 to collapse to a singlet. Irradiation experiments show that the proton at τ 7.12 is not coupled to the one at τ 8.30, but that both of them are coupled to the same region. Irradiation at τ 7.62 collapses the τ 7.12 and 8.30 to simple doublets, J = 11 cps, and strong irradiation in the τ 7.83 region collapses the τ 8.30 signal to a sharp singlet and the τ 7.12 signal to a singlet with a shoulder.

Nuclear magnetic resonance spectrum in perdeuteroacetic acid: τ 3.74 (sharp singlet, IH), 6.15 (broad, IH), 6.36 (doublet, J = 13 cps, IH), 6.61 (doublet of doublet, J = 13 & 4 cps, IH) 7.00 (singlet, N-methyl), 8.96 (doublet, J = 6 cps, C-methyl). Ultraviolet spectrum: λ_{max}^{EtOH}, 205 mμ (ϵ= 3280); 242 (ϵ= 10,660). Mass spectrum: m/e 205 (molecular ion, 61), 204 (9), 190 (5), 177 (7), 163 (100), and 162 (16). The molecular ion at 205 indicates that dehydroluciduline has the molecular composition $C_{13}H_{19}NO$.

CATALYTIC HYDROGENATION OF DEHYDROLUCIDULINE

Dehydroluciduline (16 mg) in methanol (5 ml) was hydrogenated at atmospheric pressure over Adam's catalyst (platinum oxide, 10 mg) for three days. The catalyst was removed by filtration and the solvent removed under reduced pressure. The product, 10 mg., was purified by sublimation. The sublimate consisted of a mixture of solid and liquid

material. It showed one slightly elongated spot on t.l.c. with the same R_f value as dihydroluciduline. Infrared spectrum: $\nu_{max}^{CHCl_3}$: 3590 (free OH), 3400 (broad, -OH, hydrogenbonded), 2780 ($N-CH_3$) cm^{-1}. Weak bands present at 1720 and 1600 cm^{-1} are probably due to impurities.

Mass spectrum: m/e 209 (45), 208 (65), 194 (10), 193 (17), 192 (100), 166 (46), 165 (10), 164 (31), and 150 (17) are prominent. The t.l.c. and mass spectral fragmentation pattern indicate that the hydrogenation mixture contains dihydroluciduline and its C-2 epimer.

ATTEMPTED DEUTERIUM EXCHANGE OF DEHYDROLUCIDULINE

Dehydroluciduline (44 mg) was dissolved in a solution of deuterium chloride in acetic acid-O-d (5 ml) and allowed to stand at room temperature for 60 hours. After evaporating the solvent, a heavy water solution of anhydrous potassium carbonate was added until the solution was alkaline to litmus. The aqueous layer was extracted with chloroform. The chloroform extract was dried over anhydrous magnesium sulfate and the solvent removed under reduced pressure. The residue (18 mg) was purified by molecular distillation. The distilled product was identical with the starting material (infrared and mass spectrum).

O-p-BROMOBENZOYLDIHYDROLUCIDULINE.

p-Bromobenzoyl chloride was prepared according to the method described by Vogel(94) and was used without further purification. p-Bromobenzoic acid (400 mg) was refluxed with excess thionyl chloride (4 ml) for one hour. The reflux condenser was replaced with a guard tube filled with cotton wool and "Drierite" and the mixture was kept at 120°C for two hours. After flushing the flask containing p-bromobenzoyl chloride with nitrogen several times, dihydrolucidulline (72 mg) in pyridine (3 ml) was added to the flask containing p-bromobenzoylchloride. The mixture was kept at 120° for several hours.

The cooled reaction mixture was diluted with water and acidified with dilute hydrochloric acid. The acid solution was extracted several times with diethyl ether. The aqueous solution was basified with aqueous ammonium hydroxide and extracted with chloroform. The chloroform solution was dried over anhydrous sodium sulfate. Removal of chloroform under reduced pressure gave 0-p-bromobenzoyldihydroluciduline (21 mg) as a crystalline solid. It was recrystallized from 95% ethanol, m.p. 72-73°.

Density of crystals: 1.365 g/cm.^{3} (by flotation in aqueous potassium iodide). Infrared spectrum $\nu_{max}^{CHCl_3}$, 2780 (N-CH_3), 1715 (ester carbonyl) 1600, 1270 cm^{-1}.

Nuclear magnetic resonance spectrum in $CDCl_3$: τ 2.26 (AA'BB' quartet, J = 8 cps, 4H), 4.80 (multiplet, $W_{\frac{1}{2}h}$ = 24 cps; IH), 6.80 (doublet, J = 12 cps; IH) 7.30 (multiplet, 2H), 7.82 (singlet, N-methyl), 9.10 (doublet, J = 6 cps, C-methyl).

Mass spectrum: m/e 393 (1), 392 (1), 391 (1), 390 (0.8), 350 (1), 348 (1), 193 (14), 192 (100), 185 (2), 183 (2), 164 (1), 157 (1), 155 (1), 150 (2), and 44 (14). Exact mass of the molecular ion shows that O-p-bromobenzoyl-dihydroluciduline has the molecular formula $C_{20}H_{26}NOBr$ (Calculated: 391. 1148. Found:391.1153).

LUCIDULINE LACTAM.

Potassium permanganate (1.200 g) was added in portions to luciduline (558 mg) in 20 ml of acetone freshly distilled from potassium permanganate. After four hours, the mixture was acidified with dilute hydrochloric acid (45-50 ml) and then extracted continuously with ether for 35 hours. The ether extract was dried over anhydrous magnesium sulfate. Removal of the ether under reduced pressure gave the luciduline lactam (67 mg; one spot on t.l.c.). Further purification was effected by filtration through alumina, followed by molecular distillation.

Extraction of the basified aqueous layer gave, after the usual procedure, 418 mg of pure luciduline (t.l.c. and infrared spectrum).

In some later experiments, carried out under similar conditions, no starting material could be recovered and extraction of the acidified reaction mixture afforded a large amount of non-basic reaction products. However, chromatographic filtration over alumina afforded the amounts of the ketolactam indicated in the table below:

Experiment	1	2
Weight of luciduline used (mg)	418	639
Total weight of non-basic products (mg)	300	269
Amount ketolactam isolated (mg)	40	64

The acidic portion of the non-basic oxidation products was not investigated.

Luciduline lactam was obtained as an oil which partly solidified after a few hours in the fridge. It has the molecular formula $C_{13}H_{19}NO_2$. (See mass spectral data on page 126). Infrared spectrum: $\nu_{max}^{CHCl_3}$, 1730 cm^{-1} (ketone carbonyl), 1640 cm^{-1} (lactam carbonyl).

Nuclear magnetic resonance spectrum in $CDCl_3$: τ 6.36 (quartet, IH, coupled to τ 8.64 and τ 7.92), 6.72 (multiplet, IH, coupled to τ 7.90, τ7.71 and τ7.63, 7.04 (singlet,

N-methyl), 9.05 (doublet, J = 6 cps, C-methyl).

Mass spectrum: m/e 221 (100, molecular ion), 193 (35), 180 (27), 178 (25), 165 (45), 151 (34), 149 (39), 136 (22), 111 (30), 110 (80), 108 (39), 95 (37), 94 (70), 68 (35), 57 (50), 55 (71) and 41 (78).

The exact mass determinations are tabulated below:

Molecular composition	Calculated	Found
$C_{13}H_{19}NO_2$	221.1416	221.1413
C_6H_8NO	110.0606	110.0605
C_6H_8N	94.0659	94.0659
C_4H_7	55.0548	55.0550
C_3H_5N	55.0422	55.0425
C_3H_3O	55.0184	55.0186

DEHYDROLUCIDULINE LACTAM

Freshly sublimed selenium dioxide (58 mg.) was added to luciduline lactam (116 mg) in dioxane (17 ml) and the mixture refluxed for 18 hours. After the usual work-up (see Dehydroluciduline above), dehydroluciduline lactam was obtained as a crystalline solid embedded in a persistent and oily, probably hydrocarbon, impurity. At this stage, t.l.c. analysis indicated the presence of three

components, only one of which was positive to Dragendorff's reagent. Some of the impurities were removed by filtering through alumina. The dehydroluciduline lactam so obtained weighed 35 mg and was purified further by molecular distillation under vacuum. Infrared spectrum: $\nu_{max}^{CHCl_3}$, 1685 (ketone carbonyl), 1640 (amide carbonyl) cm.$^{-1}$ Ultraviolet spectrum: λ_{max}^{EtOH}, 206 mμ, 230 mμ, 255 mμ (shoulder).

Mass spectrum: The peak at m/e 219 analyses for $C_{13}H_{17}NO_2$ (Calculated, 219.1259. Found, 219.1260).

Nuclear magnetic resonance spectrum in chloroform: τ 4.16 (sharp singlet, IH), 6.10 (quartet, IH) 6.76 (broad, IH), 7.10 (singlet, N-methyl), τ 9.00 (doublet, J = 6 cps, C-methyl).

REDUCTION OF WEAK BASES WITH SODIUM BOROHYDRIDE

A magnetically stirred solution of a mixture of weak bases (2.65 g; from counter-current distribution tubes 30-70) in methanol (40 ml) was cooled in an ice bath. A large excess of sodium borohydride (3.80 g) was added in portions over a period of 30-60 minutes. Stirring was continued for a further 3-4 hours. The reaction mixture was diluted with ice-cold water. A copious white precipitate was formed on diluting the reaction mixture with water. Extraction with chloroform, followed by the usual work-up gave the reduction products (2.11g). T.l.c. analysis indicated the presence of two major components corresponding to dihydrolucidine-B (less polar) and dihydrolucidine-A (more polar), and three or four minor components. The mixture was separated by elution chromatography over basic alumina($\sim$70g) Elution with methylene chloride (about 2 litres) afforded fractions from which pure dihydrolucidine-B (one spot on t.l.c.) was obtained as a stiff white foam (0.710 g). Further elution with ethyl acetate gave the more polar dihydrolucidine-A (0.791 g). Dry-column chromatography was used to further purify samples of dihydrolucidine-B and dihydrolucidine-A.

In recent work, dihydrolucidine-B and dihydrolucidine-A have been conveniently prepared by the sodium borohydride reduction of lucidine-B and lucidine-A respectively.

DIHYDROLUCIDINE-B

Dihydrolucidine-B, $C_{30}H_{51}N_3O$, is optically active $[\alpha]_D + 3°(C,0.26,CHCl_3)$ and displays no Cotton Effect in its O.R.D. curve. Infrared spectrum: $\nu_{max}^{CCl_4}$, 2780 ($N-CH_3$), 1620 N-CO-) cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: τ 5.0-7.3 (minor peaks), 7.38, 7.58, 7.90, 7.92 and 7.95 ($N-CH_3$ and $N-CO-CH_3$), 8.85, 8.95 (C-methyl, doublet), 9.14 (C-methyl doublet, J=7 cps). In trifluoroacetic acid, the n.m.r. spectrum has signals at τ 7.30 (N-methyl) and 7.78 (N-acetyl). Mass spectrum (sample purified by evaporative distillation): m/e 469 (55), 468 (30), 467 (35), 275 (37), 274 (31), 273 (100), 261 (91), 260 (57), 166 (77), 165 (58), 164 (45), 97 (44), 96 (54). (See Fig. 18). The following exact masses have been determined:

Molecular Formula	Calculated	Found
$C_{30}H_{51}N_3O$	469.4032	469.4019
$C_{18}H_{29}N_2$	273.2331	273.2335
$C_{17}H_{28}N_2$	260.2257	260.2252

Titration of dihydrolucidine-B with hydrochloric acid in methyl cellosolve gave no inflexion in the titration curve. Neutralization equivalent of dihydrolucidine-B = 233. An acetic acid solution of dihydrolucidine-B was titrated with perchloric acid by J.F. Alicino. Thus dihydrolucidine-B has two basic nitrogens.

DIHYDROLUCIDINE-A

Dihydrolucidine-A is a white amorphous solid. Infrared spectrum: $\nu_{max}^{CCl_4}$, 2780 (N-CH_3), 1620 (amide $>$C=O) cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: τ 5.20-7.70(minor peaks), 7.91 and 7.95 (N-CH_3 and N-CO-CH_3); these peaks undergo a downfield shift to τ 7.25 (N-methyl) and τ 7.75 (N-acetyl) in trifluoroacetic acid); τ 9.14 (CH_3-C- multiplet). Mass spectrum: 469 (18), 468 (14), 467 (17), 275 (22), 274 (21), 273 (87), 262 (21), 261 (100), 166 (24), 164 (17) (See Fig. 19). The molecular ion at m/e 469, indicates that dihydrolucidine-A has the molecular formula $C_{30}H_{51}N_3O$.

DESACETYLUCIDINE-B

Lucidine-B (206 mg) was refluxed with aqueous sulfuric acid (10% by volume) for 72 hours. The reaction mixture was diluted with ice water and basified with ammonium hydroxide and then extracted with chloroform. The chloroform extract was dried over anhydrous sodium sulfate. Removal of chloroform under reduced pressure gave desacetyllucidine-B as a slightly yellowish white solid (98 mg). Crude desacetyllucidine-B had $[\alpha]_D$-51° (C,0.18, chloroform). The sublimed sample has $[\alpha]_D$- 71°(C,0.10, chloroform). The optical rotatory dispersion curve has a single negative Cotton Effect. Infrared spectrum:$\nu_{max}^{CCl_4}$, 2779 (N-methyl), 1702, 1645 ($>$C=N-) cm^{-1}. The absorption at 1702 cm^{-1} seems to be due to the presence of an impurity. Nuclear magnetic resonance in $CDCl_3$ (100 Mcs): No peaks below τ 6.50; τ 6.8

(multiplet), 7.30, 7.84 (N-methyl singlet), 8.96 (doublet, J = 7 cps, C-methyl), 9.04(doublet, J = 6 cps, C-methyl). Mass spectrum: 425 (35, molecular ion), 273 (100),260 (51). The molecular ion corresponds to the molecular formula $C_{28}H_{47}N_3$.

DESACETYLDIHYDROLUCIDINE-B

Dihydrolucidine-B (500 mg) was refluxed with aqueous sulfuric acid (10% by volume) for 80 hours. The reaction mixture was diluted with cold water, basified with ammonium hydroxide and extracted with chloroform. The chloroform extract was dried over anhydrous sodium sulfate. Removal of the solvent under reduced pressure gave desacetyldihydrolucidine B (366 mg) as a white foamy solid. Desacetyldihydrolucidine-B has the molecular formula $C_{28}H_{49}N_3$. Calculated, 427.3926 Found, 427.3922.

Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2780 ($N-CH_3$), 1460, 1440 cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: No peaks below τ 6.50; τ 6.8-7.70 (minor peaks), 7.90 (N-methyl, shifts to τ 7.33 in trifluoroacetic acid), 9.00 (methyl doublet, J = 6 cps), 9.18 (methyl, doublet, J = 6 cps). Mass spectrum: m/e 427 (38), 275 (19), 273 (14), 262 (27), 261 (100), 166 (29), 165 (29), 164 (33), 150 (28), 110 (19). (See Fig.21).

The molecular composition of some of the peaks are tabulated on the following page.

Molecular Composition	Calculated	Found
$C_{18}H_{29}N_2$	273.2330	273.2326
$C_{17}H_{29}N_2$	261.2330	261.2324
$C_{11}H_{18}N$	164.1439	164.1444
$C_{10}H_{16}N$	150.1283	150.1281

Kuhn-Roth oxidation. Calculated for one C-CH_3, 3.51%. Found, 3.90%. Titration of desacetyldihydrolucidine-B with hydrochloric acid: no inflexion in the titration curve.

N-ACETYLLUCIDINE-B

Lucidine-B (140 mg) was dissolved in a mixture of pyridine (2 ml) and acetic anhydride (3 ml). After 12 hours, the solvents were evaporated under reduced pressure and the residue basified with saturated sodium bicarbonate, and then extracted with chloroform. The chloroform extract was dried over anhydrous sodium sulfate. Removal of the solvent gave a yellowish white solid (108 mg). Mass spectrum: m/e 509 (100), 465 (24), and a broad metastable peak centred at m/e 427 corresponding to the fragmentation m/e 509 → m/e 465. Mass spectrometric analysis: Calculated for $C_{30}H_{51}N_3O_2$, 509.3981. Found, 509.3967. Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2780, 1625 cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: τ7.88, 7.91, 7.93 (N-CH_2 and N-CO-CH_3); τ 8.72, 8.87, 8.98, 9.11 (CH_3 doublet J = 6 cps).

TETRAHYDRODEOXYLUCIDINE-B

Dihydrolucidine-B (0.794 g) in anhydrous tetrahydrofuran (100 ml) was refluxed for 24 hours with an excess of lithium aluminium hydride (1.100 g). The product was isolated according to the method described by Micovic and Mihailovic (61). The reaction mixture was cooled in an ice-bath and excess lithium aluminium hydride was decomposed by the addition of water (1.1 ml), 15% aqueous sodium hydroxide (1.1ml), and water (3.3ml) to the vigorously stirred mixture. The mixture was stirred for another 30 minutes and then filtered under section. The solvent was evaporated under reduced pressure. The residue was dissolved in dilute hydrochloric acid (10% by volume) and after extracting with ether to remove non-basic impurities, it was basified with sodium hydroxide (15% by volume) and extracted with chloroform. The chloroform extract was dried over anhydrous sodium sulfate and the solvent evaporated. This procedure gave tetrahydrodeoxylucidine-B (0.522 g) as a white solid (foam). Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2780, 1455 cm^{-1}. Nuclear magnetic resonance spectrum in deuterochloroform: No peaks below τ 6.70; τ 7.0 (broad peak), 7.50(unassigned signals plus CH_2 quartet, J = 7.5 cps), coupled to τ 8.93), 7.89 (N-CH_3, singlet), 8.93 (methyl triplet, J = 7.5 cps coupled to τ 7.50; methyl doublet, J = 7.5 cps, coupled to τ 7.83 region), 9.17 (methyl doublet, J = 6 cps, coupled to τ 8.35 region). Analysis. Calculated for $C_{30}H_{53}N_3$, C,79.06; H 11.72; N, 9.22. Found, C, 78.34;

H 11.54, 11.42; N 9.18, 8.62. Mass spectrum: (Fig.22) m/e 455 (80), 454 (32) 440 (11), 426 (24) 275 (44), 273 (34), 261 (100), 180 (64), 178 (62), and 164 (32). In an earlier mass spectrum m/e 180 was the base peak. The high resolution data for m/e 455, 261 and 180 are summarized in the table below.

Formula	Calculated	Found
$C_{30}H_{53}N_3$	455.4250	455.4237
$C_{17}H_{29}N_2$	261.2331	261.2329
$C_{12}H_{22}N$	180.1752	180.1755

ACETYLATION OF DIHYDROLUCIDINE-B

Dihydrolucidine-B (96 mg) was dissolved in a 2:1 acetic-anhydride-pyridine mixture (3 ml). After twelve hours the solvent was evaporated and the product isolated according to the method described above. N-Acetyldihydrolucidine-B (72 mg) was isolated as a slightly yellowish white solid. Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2790, 1620 cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: $_O\tau$ 4.6-7.7 (minor peaks); 7.86, 7.90, 7.92, 7.95 ($N-CH_3$ and $N-\overset{\|}{C}-CH_3$); 8.85; 8.95; 9.16 (methyl doublet, J = 6 cps). Mass spectrum: weak spectrum, peaks in the high mass range at m/e 511, 510, 509, 508, 507, 494, 468 and 466.

ACETYLATION OF TETRAHYDRODEOXYLUCIDINE-B

Tetrahydrodeoxylucidine-B (90 mg) was dissolved in a 3:1 acetic anhydride-pyridine mixture (4 ml). After 12 hours,

the mixture was worked up by the method already described. N-acetyltetrahydrodeoxylucidine-B (71 mg) was obtained as a white solid (foam). Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2790, 1620, 1615 cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: τ 6.8-7.8 (small peaks) 7.90 ($N\text{-}CH_3$ and $N\text{-}\overset{O}{\overset{\|}{C}}\text{-}CH_3$), 8.93 (methyl triplet J = 7 cps) 9.12 (C-methyl multiplet). Mass spectrum: m/e 497 (23, molecular ion), 482 (3), 454 (8), 319 (4), 317 (5), 261 (5), 181 (16), 180 (100), metastable at m/e 390 corresponds to the fragmentation m/e 497→454.

DIHYDROLUCIDINE-B-d_1

Lucidine-B (34 mg) was dissolved in methanol (10 ml) and reduced with sodium borodeuteride (30 mg) in the usual way (See reduction of weak bases, p.128). After work-up, dihydrolucidine-B (26 mg) was obtained. Mass spectrum: prominent peaks at m/e 471 (12), 470 (30), 274 (55), 273 (100), 262 (36), 261 (44), 260 (90), 259 (26). The corresponding peaks in the mass spectrum of dihydrolucidine-B taken at about the same time have the following intensities: m/e 471 (1), 470 (4), 274 (4), 273 (100), 262 (16), 261 (50), 260 (93), 259 (27). It was observed, however, that the relative intensities of the peaks in the mass spectra of dihydrolucidine-B changed from time to time even on exactly the same sample.

TETRAHYDRODEOXYLUCIDINE-B-d_3

Dihydrolucidine-B-d_1 (20 ml) was reduced with lithium

aluminium deuteride (140 mg) using the method described under the preparation of tetrahydrodeoxylucidine-B (page 133). Mass spectrum (Fig. 23): m/e 459 (39), 458 (92), 457 (53), 456 (31), 276 (37), 263 (49), 261 (33), 260 (31), 196 (39), 183 (45), 182 (100), 181 (47), 180 (67), 165 (32), 164 (36), 163 (30).

DESACETYLDIHYDROLUCIDINE-B-d_1

Desacetyllucidine-B (8 mg) was dissolved in methanol (10 ml), and reduced with excess sodium borodeuteride (50 mg) using the method already described under the preparation of dihydrolucidine-B (page 135). Desacetyldihydrolucidine-B-d_1, so obtained (7 mg) has the following prominent peaks in its mass spectrum: m/e 429 (21), 428 (58), 427 (23), 276 (20), 273 (17), 263 (31), 262 (100), 261 (19), 260 (17), 167 (19), 166 (31), 165 (35), 164 (29), 150 (24).

DESACETYLDIHYDROLUCIDINE-A

Dihydrolucidine-A (512 mg) was refluxed with dilute sulfuric acid (30 ml) for 72 hours. The product was isolated using the procedure described under desacetyldihydrolucidine-B (page 131). Desacetyldihydrolucidine-A (362 mg) was obtained as yellowish solid (foam). Infrared soectrum: $\nu_{max}^{CHCl_3}$, 2780 (N-CH_3). 1460, 1440 cm^{-1}. Nuclear magnetic resonance spectrum in $CDCl_3$: No peaks below τ 6.0; τ 6.00-7.75 (minor peaks), 7.94 (N-methyl singlet; shifts to τ 7.33 in trifluoroacetic acid), 9.17 (C-CH_3 multiplet). Mass spectrum: m/e 427 (31, molecular ion),

526 (18), 425 (21), 275 (17), 273 (14), 261 (94), 166 (30), 165 (30), 164 (45), and 150 (57) in the high mass range; and m/e 44 (100). The molecular ion at m/e 427 and other data suggest that desacetyldihydrolucidine-A has the molecular formula $C_{28}H_{49}N_3$.

TETRAHYDRODEOXYLUCIDINE-A

Dihydrolucidine-A (806 mg) was dissolved in anhydrous tetrahydrofuran (100 ml) and reduced with excess lithium aluminium hydride (1.30 g) according to the method described under tetrahydrodeoxylucidine-B. Tetrahydrodeoxylucidine-A (683 mg) was obtained as a white solid (foam). Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2780 (N-CH_3) 1460 cm^{-1}. Nuclear magnetic resonance spectrum: τ 6.35-7.10 (minor peaks), 7.50 (multiplet, includes well defined -CH_2 quartet, splitting 7.5 cps, coupled to τ 8.93), 7.91 (N-methyl, singlet), 8.93 (triplet, splitting 7.5 cps, coupled to τ 7.50), 9.15 (multiplet, C-CH_3, coupled to τ 8.30 region). There was no observable change in the spectrum after shaking with D_2O. Mass spectrum: m/e 456 (34), 455 (100), 454 (42), 453(19), 440 (8), 427 (16), 426 (46), 397 (8), 276 (10), 275 (40), 273 (11), 262 (24), 261 (92), 260 (8), 219 (10), 195 (16), 194 (24), 192 (13), 191 (8), 181 (9), 180 (30), 179 (42), 178 (94), 166 (13), 165 (18), 164 (22) and 150 (9).

DESACETYLLUCIDINE-A

Desacetyllucidine-A was prepared by acid hydrolysis of

lucidine-A according to the method already described under desacetyllucidine-B. Infrared spectrum: $\nu_{max}^{CHCl_3}$, 2780, 1710, 1645 cm^{-1}. Mass spectrum: m/e 426 (28), 425 (87, molecular ion), 424 (20),423 (13), 275 (20), 274 (28), 273 (100), 262 (13), 261 (30), 260 (60), 259 (12), 257 (19), 233 (13), 218 (28), 217 (25), 192 (17), 191 (12), 177 (14), 166 (33), 165 (13), 152 (20), 151 (20), and 150 (69). Desacetyllucidine-A displays Cotton effects in its optical rotatory dispersion curve.

DIHYDROLYCOLUCINE

Lycolucine (40 mg) in methanol (15 ml) was hydrogenated at atmospheric pressure over Adam's catalyst (platinum oxide, 20 mg) for 20 hours. The catalyst was removed by filtration and the solvent removed under reduced pressure. The product, dihydrolycolucine was obtained as a white solid (32 mg). It has a higher R_f value than lycolucine on alumina (t.l.c.). Infrared spectrum: $\nu_{max}^{CCl_4}$, 2780 (NCH_3), 1730, 1638 (amide carbonyl) 1580, 1560, 1460, 1450 cm^{-1}. Ultraviolet absorption spectrum: λ_{max}^{EtOH}, 216 mμ (intense), 275 mμ (less intense than 216 mμ band). On acidification with dilute hydrochloric acid, the intensity of the 275 mμ band increases whereas the intensity of the 216 mμ band diminishes slightly. Nuclear magnetic resonance spectrum in $CDCl_3$: τ 2.70, 2.78, 2.96, 3.22 and 3.30 (complex set of peaks, aromatic protons), 5.0-7.6, 7.86 (CH_3-N), 7.96 (N-CO-CH_3), 8.77, 9.10(CH_3-C, complex

multiplet). Mass spectrum: m/e 464(14), 463 (33, molecular ion), 458 (15), 420 (18), 273 (14), 271 (24), 270 (100), 150 (15), metastable peak at m/e 381 corresponds to m/e 463→420 (Calculated m/e 382).

DESACETYLDIHYDROLYCOLUCINE

Dihydrolycolucine (32 mg) was refluxed with 15% sulfuric acid by volume, 20 ml) for 3 days. After the usual work-up, desacetyldihydrolycolucine (26 mg) was obtained as a solid (foam). Infrared spectrum: $\nu_{max}^{CCl_4}$, 2780 (N-CH_3), 1580 cm^{-1}. Ultraviolet spectrum: λ_{max}^{EtOH}, 215 mμ (ϵ=8500), 275mμ (ϵ =6300). Nuclear magnetic resonance spectrum in $CDCl_3$: τ 3.04 (quartet, splitting 8 cps), 7.90 (N-CH_3), 8.7, 9.10 (CH_3-C, multiplet). Mass spectrum: m/e 422 (17), 421 (55, molecular ion), 420 (14), 297 (12), 271 (30), 270 (100), 268 (11), 261 (10), 258 (16), 257 (67), 185 (21), 164 (11), 152 (20), 150 (48), and 149 (10).

OCTAHYDRODESACETYLLYCOLUCINE

Desacetyldihydrolycolucine (20 mg) was dissolved in isoamyl alcohol (25 ml) and heated under reflux in an oil bath kept at about 140°. Excess sodium (1.0 g) was added in small portions over a thirty minute period and the mixture was refluxed for a further 5 hours. The cooled reaction mixture was diluted with water and then acidified with dilute aqueous sulfuric acid. The acidified solution was extracted with ether and the ether extract discarded. The aqueous solution was basified and

extracted with chloroform. The chloroform solution was dried over anhydrous magnesium sulphate and the solvent evaporated under reduced pressure. Crude octahydrodesacetyllycolucine (18 mg) was obtained as a solid (foam) and purified by evaporative distillation. Mass spectrum: m/e 428 (12), 427 (35, molecular ion), 426 (23), 275 (24), 274 (9), 273 (20), 262 (23), 261 (100), 260 (13), 218 (10), 166 (17), 165 (14), 164 (21) and 150 (24). T.l.c. analysis showed two major and several minor spots; none of the major spots corresponded to desacetyldihydrolucidine-A, two of the minor spots have very nearly the same R_f value as desacetyldihydrolucidine-A.

DESACETYLLYCOLUCINE

Lycolucine (140 mg) was dissolved in 10% sulfuric acid and refluxed for three days. Desacetyllycolucine (94 mg) was obtained as a white solid using the isolation procedure described under desacetyllucidine-B (page 130). Infrared spectrum: $\nu_{max}^{CCl_4}$, 2778 (N-CH_3), 1650 (C=N), 1580, 1455, 1370 cm^{-1}. Ultraviolet spectrum: λ_{max}^{EtOH} : 218, 262, 300 mμ; very similar to lycolucine. On acidification, 215 mμ (less intense than 218 mμ above), 261 mμ (less intense than 262 mμ above), 320 mμ (more intense than 300 mμ above). Optical rotation: $[\alpha]_D$ + 184° (C, 0.23, chloroform). Nuclear magnetic resonance spectrum in $CDCl_3$: τ 2.80 and 3.30 (quartet, J = 7 cps, 2H), 3.40 (singlet,IH), 6.5-7.65 (several peaks, about 14H), 7.90 (N-CH_3, singlet), and 9.06 (CH_3-C, multiplet, about 12H).

Nuclear magnetic resonance spectrum in perdeuteroacetoc acid: τ 1.78 and 2.60 (quartet, J = 7 cps, 2H), 3.04 (singlet, 1H), 7.10 (N-CH_3, singlet), 9.00 (CH_3-C, multiplet). Mass spectrum: m/e 419 (24, molecular ion), 270 (36), 269 (30), 268 (86), 256 (13), 255 (53), 164 (9), 152 (14), 151 (15), 150 (100), and 98 (23). The following exact masses were determined:

Molecular Formula	Calculated	Found
$C_{28}H_{49}N_3$	419.330	419.3298
$C_{10}H_{16}N$	150.1283	150.1284

SELENIUM DEHYDROGENATION OF DIHYDROLUCIDINE-B

Dihydrolucidine-B (200 mg) was mixed with selenium (800 mg) in a thick-walled Carius tube and then sealed under vacuum and heated at 290-300° for about 20 hours. The cooled Carius tube was opened, placed inside a larger tube and the dehydrogenation products were isolated by evaporative distillation under vacuum. The total distillate (115 mg) was dissolved in benzene (20ml) and extracted four times with 20 ml portions of dilute hydrochloric acid (10% by volume). The benzene solution was dried over anhydrous sodium sulfate and on removal of the solvent under reduced pressure had a viscous mixture of non-basic compounds (25 mg). The aqueous extract was basified with aqueous ammonium hydroxide. Extraction with methylene chloride afforded the basic products (56 mg).

Gas-liquid chromatography of the basic dehydrogenation products on a column (5' x ¼") packed with 20% SE-30 on Chromosorb W, 60-80 mesh, at 202° and a helium flow rate of 35 ml/min. revealed the presence of at least twenty-two components. The mass spectrum of a sample from the first peaks had prominent peaks at m/e 143 and 157 indicative of substituted quinolines or isoquinolines. The ultraviolet spectra of components 17 and 18 (17th and 18th peaks on the gas chromatogram) suggested the presence of 2,3,6-trisubstituted pyridines (75).

THE SEPARATION AND ISOLATION OF THE DEHYDROGENATION PRODUCTS

(i) Separation into Basic, Weakly Basic and Non-Basic Fractions

The reaction flask or the Carius tube was crushed into small pieces which were transferred into a thimble of a Soxhlet extractor. The crude mixture was then extracted with methylene chloride for 4 to 8 hours. The methylene chloride was removed at the pump and the crude extract was dissolved in benzene (10-20 ml). The benzene solution was extracted several times with dilute hydrochloric acid (10% by volume). The benzene solution was dried over anhydrous sodium sulfate and the solvent removed under reduced pressure in order to obtain the non-basic products.

The aqueous solution was basified with ammonium hydroxide and extracted with chloroform. The chloroform solution was

dried over anhydrous sodium sulfate. Removal of the solvent afforded the total basic products. The basic material was dissolved in methylene chloride (about 20 ml) and then extracted several times with dilute hydrochloric acid (10% by volume). The methylene chloride solution gave the weakly basic components on removal of the solvent. Basification of the aqueous phase followed by extraction with chloroform gave the strongly basic components.

(ii) Isolation of GC-17 and GC-18

The mixture of weakly basic dehydrogenation products was chromatographed over basic alumina (BDH alumina, Activity II-III). The fraction eluted with hexane-benzene (1:1) or pentane-benzene (1:1) consisted mainly of GC-17 and GC-18 (t.l.c., g.l.c. and mass spectra). Further elution with benzene and methylene chloride gave mixtures whose ultraviolet spectra have suggestive of alkylquinolines.

The fraction containing GC-17 and GC-18 was usually further purified by sublimation and the final separation was accomplished by gas-liquid chromatography on a column 16' x ¼", 15% Apiezon L on Chromosorb W 60-80, at 240° and a helium flow rate of 60 ml/min. Under these conditions the retention times of GC-17 and GC-18 were 38 and 44 minutes respectively.

GC-17 and GC-18 were collected by passing the effluent gases through a flask containing gently refluxing methylene chloride. The flask was fitted with an air condenser partly packed with glass wool.

Recently the dehydrogenation of the dihydrolucidines (1.39 g) gave 14 mg of GC-17 and 22 mg of GC-18.

GC-17

GC-17 was obtained as an oil. Infrared spectrum: $\nu_{max}^{CCl_4}$: 1580, 1565, 1460, 1450 and 1380 cm^{-1}. Ultraviolet spectrum λ_{max}^{EtOH}, 224, 273 mμ; on acidification the 273 mμ peak is shifted to 283 mμ. Nuclear magnetic resonance spectrum in $CDCl_3$: main peaks at τ 2.96 (quartet, J = 9 cps; 2H), 7.10 (multiplet), 7.52 (singlet, $ArCH_3$) 8.70 (doublet, J = 8, isopropyl methyl groups; coupled to τ 7.03) 9.10 (CH_3-C multiplet, partly coupled to τ 8.65 region). Mass spectrum: m/e 243 (39, molecular ion), 242 (15), 229 (13), 228 (65), 186 (18), 160 (14), 158 (11), 148 (11), 147 (12), 146 (17), 136 (13), 135 (100), 134 (23), 133 (17), 132 (22), and 121 (10); a metastable peak at m/e 75.0 corresponds to the fragmentation m/e 243 → 135 (Calculated, 75.0). The mass spectrum and ultraviolet spectrum of GC-17 are very similar to GC-18.

GC-18

GC-18 is a white crystalline solid, optical rotation $[\alpha]_D$ -1° (chloroform), m.p. 85-100°. Infrared spectrum: $\nu_{max}^{CCl_4}$, 3040 - 2800, 1582, 1570, 1460, 1450, 1440, 1390, 1370, 1365 cm^{-1}. Ultraviolet spectrum: λ_{max}^{EtOH} : 221 mμ (ϵ = 4700), 272 mμ (ϵ = 5720), 278 shoulder (ϵ = 5090). Nuclear magnetic resonance spectrum in $CDCl_3$: τ 2.91 (AB quartet, J = 8 cps, 2H), 6.80 - 7.50 (multiplet, ca.3H), 7.45 (CH_3 singlet, $ArCH_3$), 8.63 (doublet, J = 7 cps, isopropyl group, coupled to τ 7.01 region),

9.00-9.10 (CH_3-C, multiplet coupled to τ 8.54 and 8.02). Nuclear magnetic resonance in perdeuteroacetic acid: τ2.19 (quartet, J = 9 cps, 2H), 6.88 (multiplet, Ar-CH_2 and Ar CH-), 7.31 (Ar-CH_2, singlet), 8.58 (doublet, J= 6 cps, isopropyl group), 9.10 (CH_3-C, multiplet). Mass spectrum (Fig. 28): m/e 243 (51, molecular ion), 242 (13), 228 (48), 186 (13), 160 (12), 146 (10), 136 (11), 135 (100), 134 (19), 133 (15), and 132 (14); metastable at m/e 75 corresponds to the fragmentation m/e 243 → 135 (calculated, 75.0). GC-18 has the molecular formula $C_{17}H_{25}N$. (Calculated, 243.1987; Found 243.1985). The peak at m/e 135 has the molecular composition $C_9H_{13}N$ (Calculated for $C_9H_{13}N$, 135.1048. Found, 135.1046).

iii. Countercurrent distribution of Strongly Basic Dehydrogenation Products

The strongly basic dehydrogenation products were dissolved in methylene chloride (25 ml) and subjected to counter-current distribution over nine funnels with aqueous potassium dihydrogen phosphate (ca 0.2M; pH 4.5) as the stationary phase. Equal volumes of each phase (25 ml) were used. In one experiment crude strongly basic compounds (1.0 g) gave the results summarized below:

Funnel	1	2	3	4	5	6	7	8	9
Amount (mg)	65	32	7	14	3	24	35	112	547

The major components in the various fractions were separated by gas-liquid chromatography.

(iv) Isolation of 7-methyldecahydroquinoline

Gas-liquid chromatography of the contents of funnel 1 gave almost exclusively one component which has been identified as 7-methyldecahydroquinoline. On a column 5' x ¼" packed with 10% QF - 1, on Chromosorb W, (60 - 80 mesh) at 130° C and a helium flow rate of 28 ml/min it has a retention time of 3.5 min. It was isolated as a white crystalline solid. Infrared spectrum: $\lambda_{max}^{CCl_4}$, 2783 (sharp), 1450 1330 cm^{-1}. Mass spectrum: (Fig.25): m/e 153 (12, molecular ion), 152 (18), 110 (7), 96 (100), and the metastable peak at m/e 60.2 corresponds to the fragmentation 153 → 96. Exact mass determination showed that the molecular formula of the dehydrogenation product is $C_{10}H_{19}N$ (Calculated, 153.1517) Found, 153.1517). The m/e 96 ion has the molecular composition $C_6H_{10}N$ (Calculated, 96.0813. Found, 96.0811). Nuclear magnetic resonance in $CDCl_3$ (on a small sample): τ 9.02 (methyl doublet, J = 6 cps).

Acetylation using the pyridine-acetic anhydride method gave an N-acetyl compound. The product was purified by evaporative distillation. Mass spectrum (Fig. 26): m/e 195 (31, molecular ion), 194 (3), 180 (12), 152 (15), 132 (60), 96 (100); metastable peaks at m/e 97.6 and 66.9 correspond to the fragmentations m/e 195 → 138 and m/e 138 → 96 respectively.

(v) Isolation of 7-methylquinoline, 7-methyl-5,6,7,8-tetrahydroquinoline and a dimethylquinoline

Gas-liquid chromatography of the contents of funnels 6 to 9 furnished varying amounts of 7-methyl-5,6,7,8,8-tetrahydroquinoline (A), 7-methylquinoline (B) and a dimethylquinoline (C) and other uncharacterized compounds. G.l.c. analysis on 10% QF - 1, on Chromosorb W, 60 - 80 mesh, (5' x ¼") at 140° and a helium flow rate of 28 ml/min, gave A, B and C with retention times, 5.0, 7.5 and 12.0 min respectively.

7-methyl-5,6,7,8-tetrahydroquinoline was obtained as a liquid. Ultraviolet spectrum: 211, 265, 268. 277 mμ; on acidification: 202, 273 mμ is more intense than the 268 mμ band). Mass spectrum (Fig. 27): m/e 147 (100, molecular ion), 146 (32), 132 (44), 105 (67). Exact mass determinations indicate that the molecular ion, M-15 and M-42 peaks have the compositions $C_{10}H_{13}N$ (Calculated, 147.1048. Found, 147.1047), $C_9H_{10}N$ (Calculated, 132.0813. Found, 132.0812), and C_7H_7N (Calculated, 105.0578. Found, 105.0582) respectively. Nuclear magnetic resonance spectrum in $CDCl_3$ on a small sample showed a doublet at τ 8.60 (J = 6 cps).

7-methylquinoline. This dehydrogenation product was isolated as a clear, colorless, mobile liquid. Infrared spectrum: $\nu_{max}^{CCl_4}$, 3020-2910, 1638, 1595, 1500 cm^{-1}; very similar to the infrared spectrum of authentic 7-methylquinoline obtained by purification of commercial 7-methylquinoline via its picrate. Ultraviolet spectrum: λ_{max}^{EtOH} : 211, 217,

240, 249, 278, 283, 305, 310, 318 mμ. The 280 mμ band disappears and 320 mμ is intensified on acidification. Mass spectrum: m/e 143 (100, molecular ion), 142 (42), and 115 (11). The exact mass of the molecular ion suggests the formula $C_{10}H_9N$ (Calculated, 143.0735. Found, 143.074).

A Dimethylquinoline

At least two dimethylquinolines have been isolated, but only the one which has been isolated in a relatively larger amount (7 mg) is described. On 15% QF - 1 column (10' x ¼") at 180° and a helium flow rate of 24 ml/min it has a retention time of 18.0 minutes. It was obtained as a white crystalline solid. Its styphnate melted sharply at 205 - 206° with decomposition. Mass spectrum: m/e 185 (3), 184 (2), 175 (6), 171 (20, 170 (7), 158 (13), 157 (100, molecular ion of a major component), 156 (36), 142 (19), 128 (9), and 115 (7). Ultraviolet spectrum: λ_{max}^{EtOH} : 210, 228, 231, 280 (broad), 310, 324 mμ. The 280 mμ band disappears and the 323 mμ is intensified on acidification. Nuclear magnetic resonance spectrum in $CDCl_3$: τ 2.18 (singlet, ca. 2H), 2.56 (AB quartet, J = 8 cps, ca. 2H), 7.46 and 7.52 (methyl singlets) τ 1.30 (broad, might be a background signal). Comparison with other n.m.r. spectra show that it cannot be any of the following: 5,7-; 2,7-; 3,7-; 3,5-; 5,6-; 6,7-; 3,6-; or 2,4-dimethylquinoline.

(vi) Non-basic Dehydrogenation Products

The non-basic extract (390 mg) was chromatographed over alumina (BDH basic alumina, 30 g, Activity II-III). Elution

with hexane (220 ml) gave a mixture (41 mg) which showed 14 peaks on g.l.c. analysis on 20% SE - 30 on Chromosorb-W (5' x ¼") at 165° and a helium flow rate of 34 ml/min. A component with retention time 10.3 min was collected and found to be identical with diphenyl (mass spectrum, g.l.c. retention time and ultraviolet spectrum). Mass spectrum: m/e 154 (100, molecular ion), 153 (27), 152 (22), 76 (12). Ultraviolet spectrum: λ_{max}^{EtOH} : 207, 246 mμ. Extensive study of the other fractions did not lead to the characterization of any other compounds.

SELENIUM DEHYDROGENATION OF LYCOLUCINE

Lycolucine (166 mg) was mixed with selenium powder (300 mg) and heated in a sealed tube at 290 - 310° for seven hours. The tube and contents were crushed and Soxhlet-extracted with methylene chloride for three hours. After removal of the methylene chloride, the total crude extract was dissolved in benzene and then extracted with dilute hydrochloric acid. The aqueous phase was basified with aqueous ammonium hydroxide and extracted with chlorform. The chloroform solution was dried over anhydrous sodium sulfate and the solvent removed under reduced pressure. This procedure gave basic dehydrogenation products (28 mg). The total crude basic extract had the following mass spectrum: m/e 243 (17), 237 (38), 236 (33), 235 (20), 234 (10), 228 (21), 222 (14), 189 (22), 157 (24), 156 (15), 148 (19), 147 (100), 144 (20), 143 (60), 142 (35), 141 (10), 135 (40), 133 (10),

132 (67), and 130 (16). The non-basic extract weighed 16 mg.

G.l.c. analysis of the basic dehydrogenation products on 10% QF - 1 on Chromosorb-W, 60 - 80 mesh (10' x ¼") at 165° with a helium flow rate of 40 ml/min, indicated the presence of one major component (more than 90% of the mixture), retention time 7.2 minutes, and several very minor components. The mass spectrum of the minor components has peaks at m/e 243 (58, molecular ion), 242 (12), 229 (14), 228 (41), 186 (17), 160 (12), 158 (11), 148 (12), 147 (13), 146 (15), 144 (13), 136 (12), 135 (100), 134 (23), 133 (20), 132 (19), and 121 (19). The mass spectra of the total crude basic products, and the minor basic dehydrogenation products indicate that the minor basic products are GC-17 and GC-18.

The major basic dehydrogenation product was shown to be identical with the pyridinoid dehydrogenation product previously identified as 7-methyl-5,6,7,8-tetrahydroquinoline as indicated by the retention time, mass spectrum and ultraviolet spectrum. Mas spectrum m/e 147 (100, molecular ion), 146 (30), 132 (60), 118 (20), and 105 (78). Ultraviolet spectrum λ_{max}^{EtOH} : 206, 268, 276 mμ; (on acidification an intense band appears at 272 mμ).

REDUCTION OF GC-18

GC-18 (12 mg) was dissolved in absolute ethanol and heated to reflux. A large excess of sodium (about 1.0g) was added in portions over 2 days to the refluxing solution. The mixture was diluted with water and then continuously extracted with ether for 6 days. The ether was evaporated and the residue was dissolved in dilute hydrochloric acid. The acidic aqueous solution was extracted with ether and the ether extracts were discarded. The aqueous solution was basified with ammonium hydroxide and then extracted with ether. The ether solution was dried over anhydrous magnesium sulfate and the solvent was evaporated under reduced pressure. Mass spectrum: the base peak at m/e 124 has the composition $C_8H_{14}N$ (calc., 124.1126; found, 124.1124).

The reduction product (about 4 mg) was dissolved in a pyridine/acetic anhydride mixture (1:1, 0.5 ml). After 10 hours, the solvents were removed under reduced pressure and the mixture basified with saturated sodium bicarbonate. The acetylated reduction product $C_{19}H_{33}NO$ was isolated in the usual way, and then purified by evaporative distallation. Mass spectrum: m/e 291 (18), 289(9), 167 (12), 166 (100), 163 (20), 124 (41), 94.6 (metastable peak) and 92.8 (metastable peak). The metastable peaks correspond to m/e 291 → 166 and m/e 166 → 124. The following exact masses were determined:

Molecular Composition.	Calculated.	Found.
$C_{19}H_{33}NO$	291.2562	291.2560
$C_{10}H_{16}NO$	166.1232	166.1229
$C_8H_{14}N$	124.1126	124.1124

PYROLYSIS OF DIHYDROLUCIDINE-A

Dihydrolucidine-A (100 mg) was pyrolysed at 310 - 320°C in a Carius tube sealed under vacuum for 16 hours. The tube and contents were crushed and Soxhlet-extracted with ether for sixty hours. The extract was separated into basic and non-basic products using the method described under the dehydrogenation of lycolucine.

The basic extract has the following ultraviolet spectrum: λ_{max}^{EtOH}, 273 mμ; on acidification, 278 mμ. The basic extract (7 mg) was analysed by gas-liquid chromatography on a column (5' x ¼") packed with 20% SE-30 on chromosorb W,60 - 80 mesh, and a helium flow rate of 30 ml/min. The chromatogram showed the presence of 3 major components GLC - 1, GLC - 2 and GLC-3 and 11 minor components. The preliminary data on the main components is summarized in the following table:

Fraction	Retention time (min)	Main peaks in mass spectrum
GLC-1	8.0 min.	m/e 147 (Base Peak), 132, 105
GLC-2	10.0 min.	m/e 161, 146 (Base Peak), 118
GLC-3	12.5 min.	m/e 161, (Base Peak), 146, 132, 119.

The ultraviolet spectrum and mass spectra suggest that mino- and di- methyl 5,6,7-8-tetrahydroquinolines are formed on pyrolysis.

The non-basic fraction (65 mg) was chromatographed on basic alumina.

The fraction eluted with hexane-benzene mixture (1:1) weighed 24 mg. On g.l.c. analysis on a column (5' x ¼") packed with 20% SE-30 on Chromosorb W (60 - 80 mesh) at 165° and a helium flow rate of 25 ml/min. afforded one major component (90%), retention time, 14.0 minutes and nine minor components.

The major component, isolated as a white crystalline solid, has the molecular formula $C_{12}H_{10}$. (Calculated for $C_{12}H_{10}$, 154.0772. Found, 154.0782. Mass spectrum: m/e 154 (100, molecular ion), 153 (27), 152 (22), and 76 (12). The Ultraviolet spectrum and mass spectrum of the pyrolysis product are identical with the ultraviolet and mass spectra of authentic diphenyl.

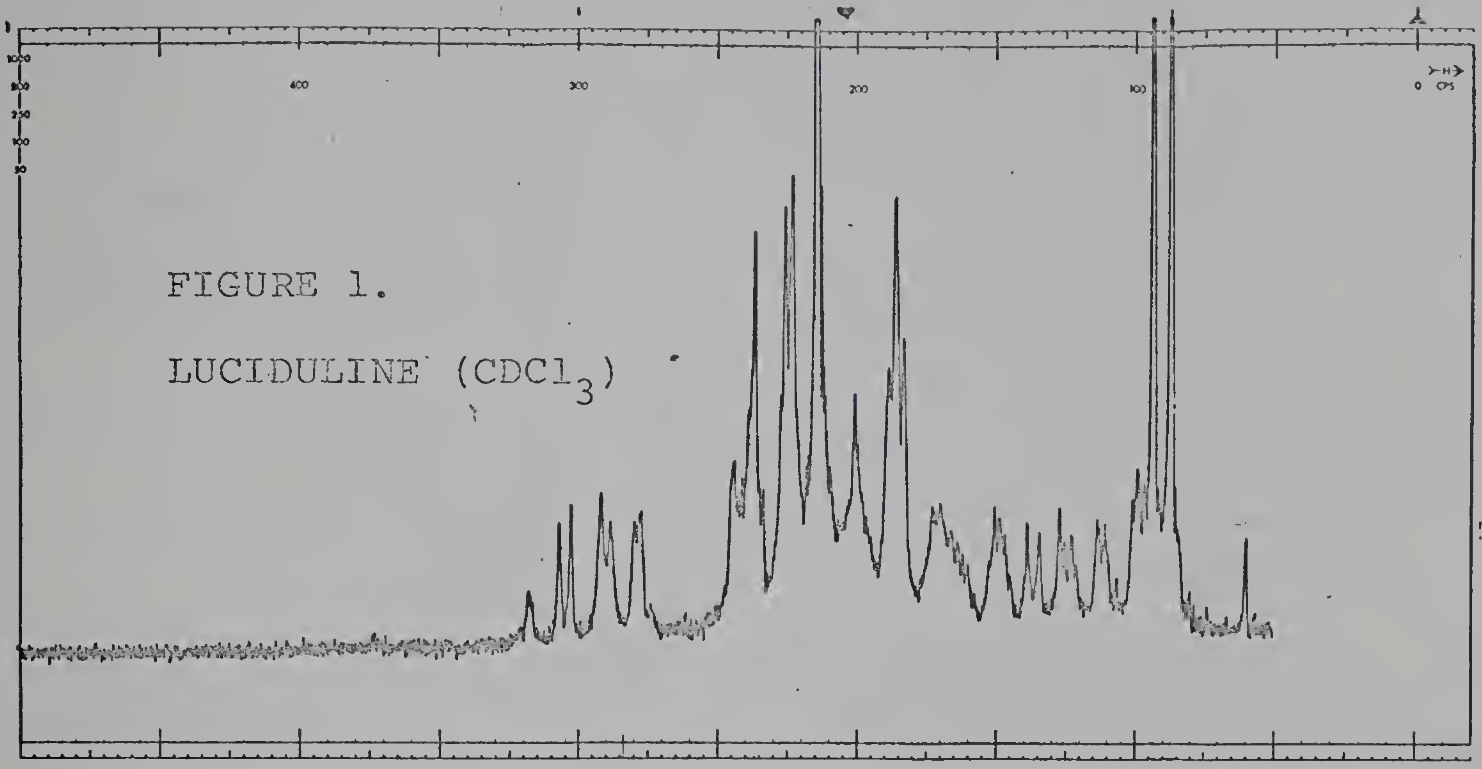


FIGURE 1.

LUCIDULINE $(CDCl_3)$

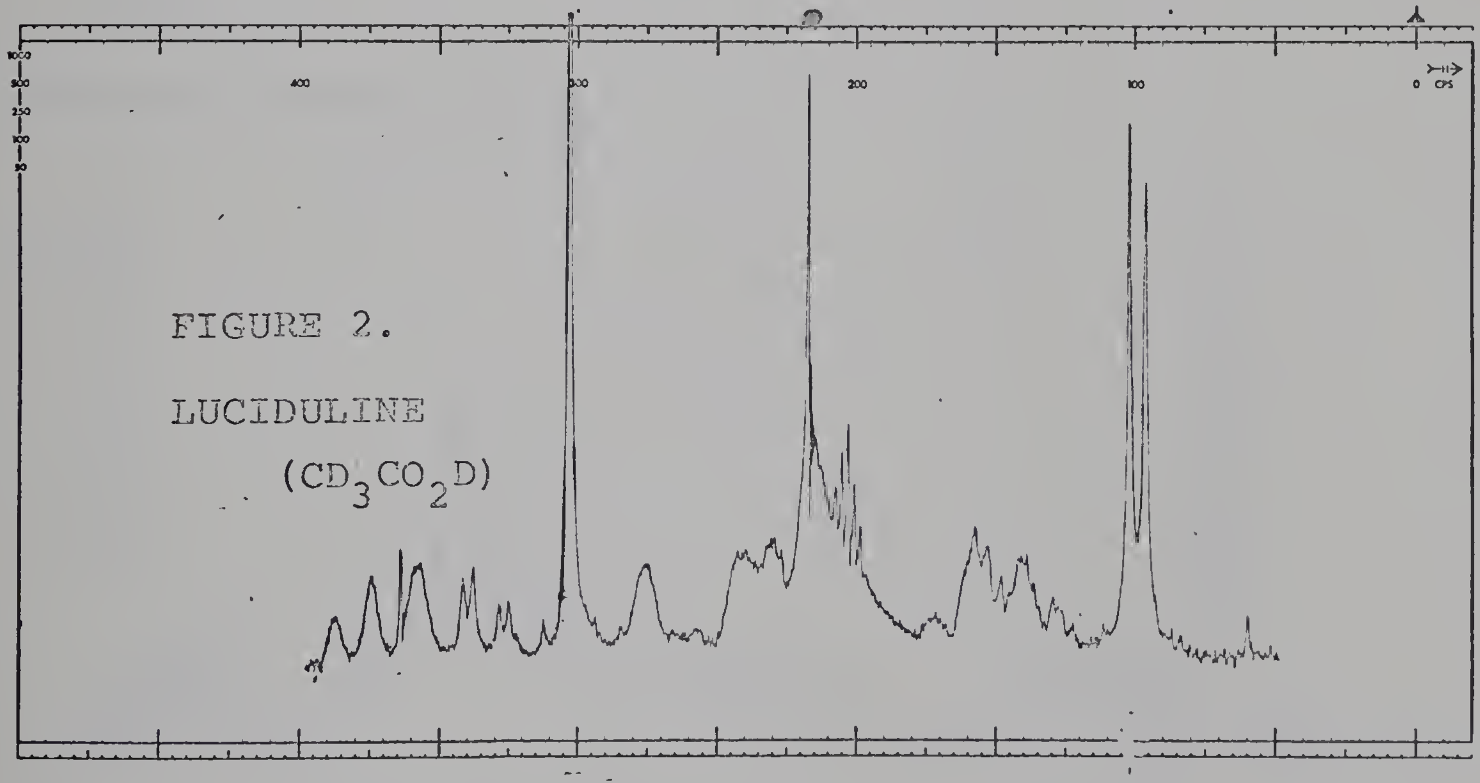


FIGURE 2.

LUCIDULINE

(CD_3CO_2D)

FIGURE 3.

LUCIDULINE-D_2 ($CDCl_3$)

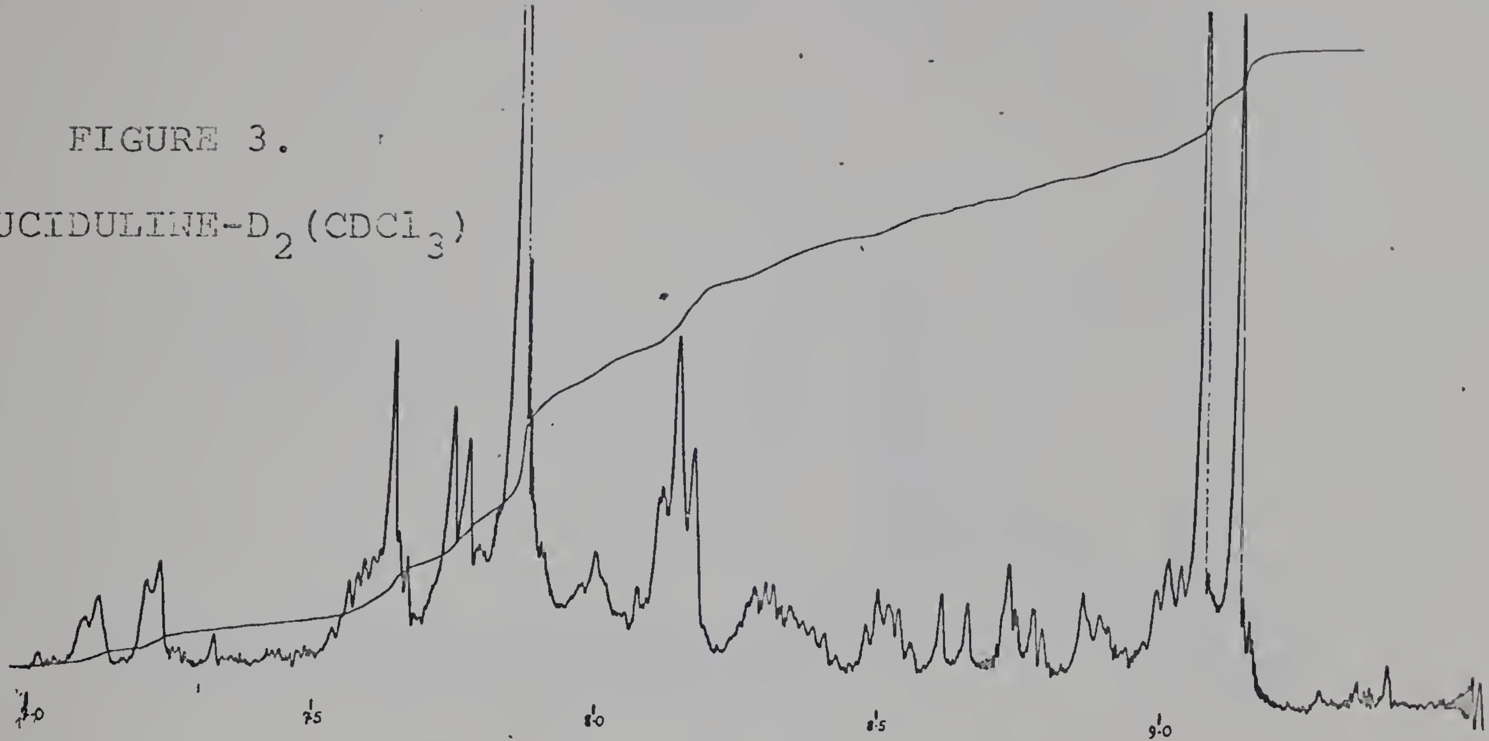


FIGURE 4.

DEHYDROLUCIDULINE ($CDCl_3$)

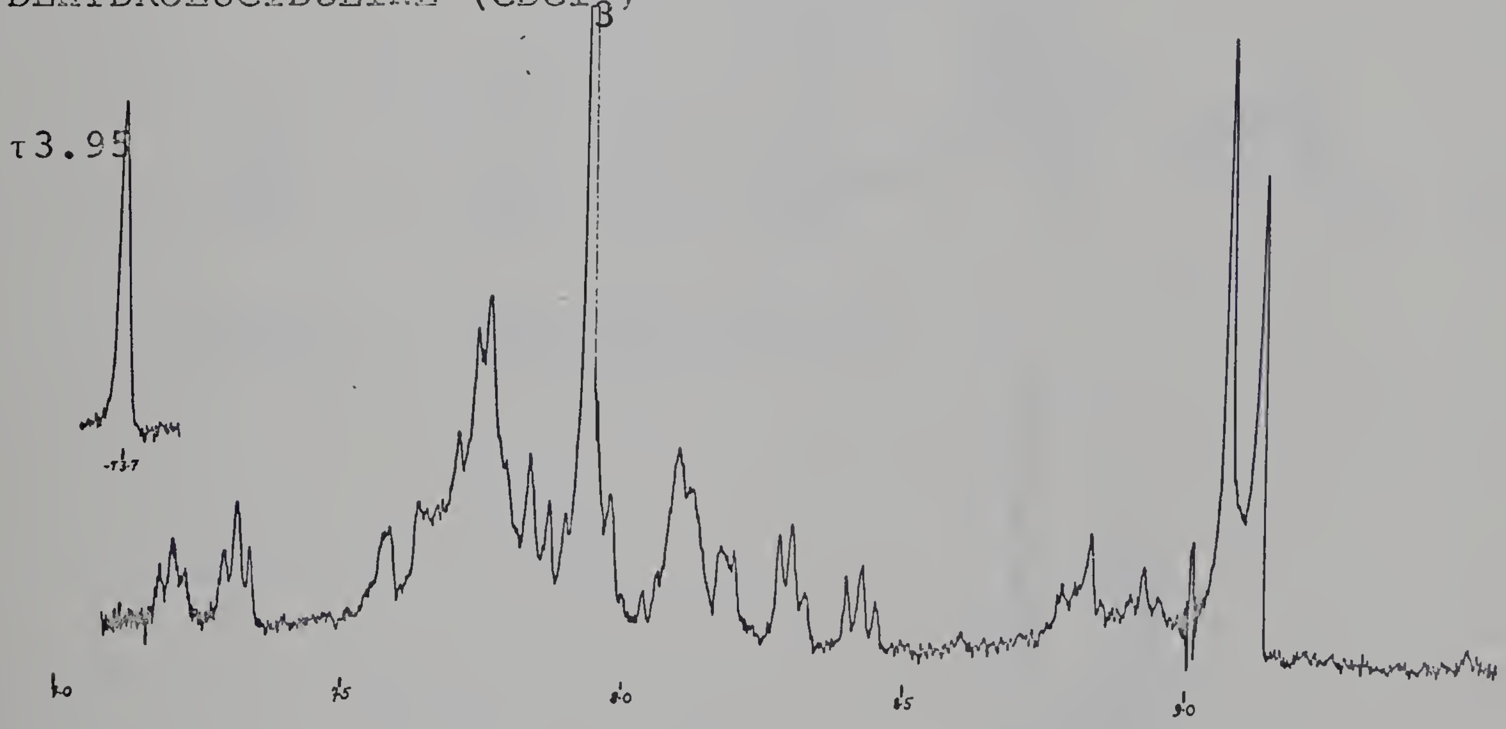
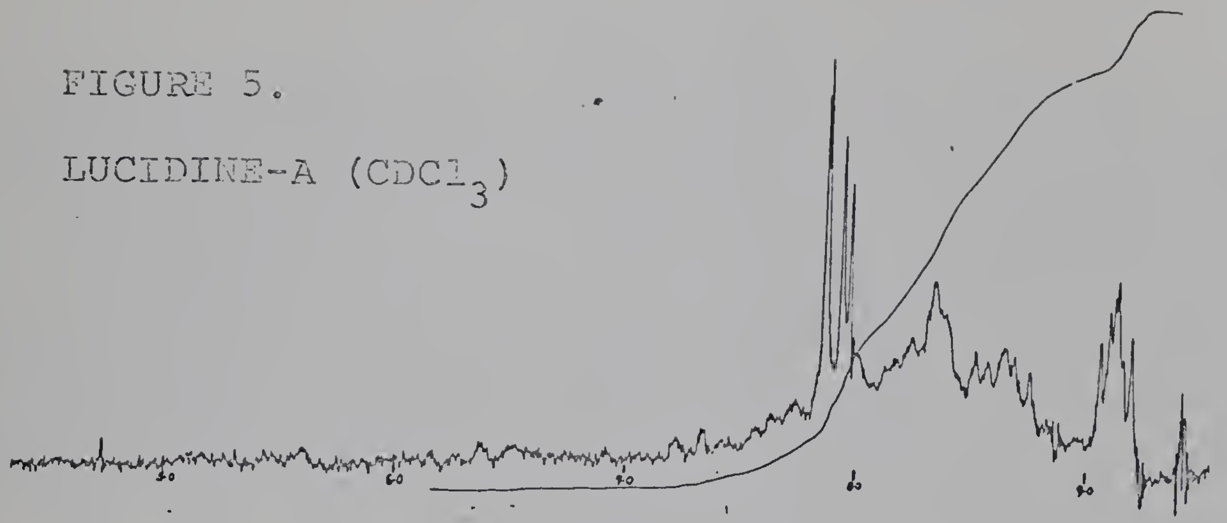


FIGURE 5.

LUCIDINE-A ($CDCl_3$)



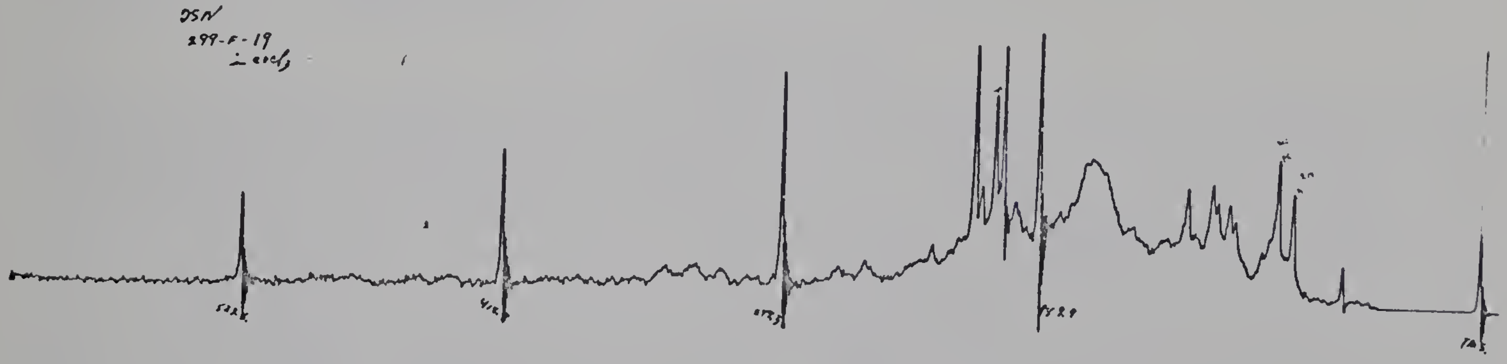


FIGURE 6. LUCIDINE-B ($CDCl_3$)

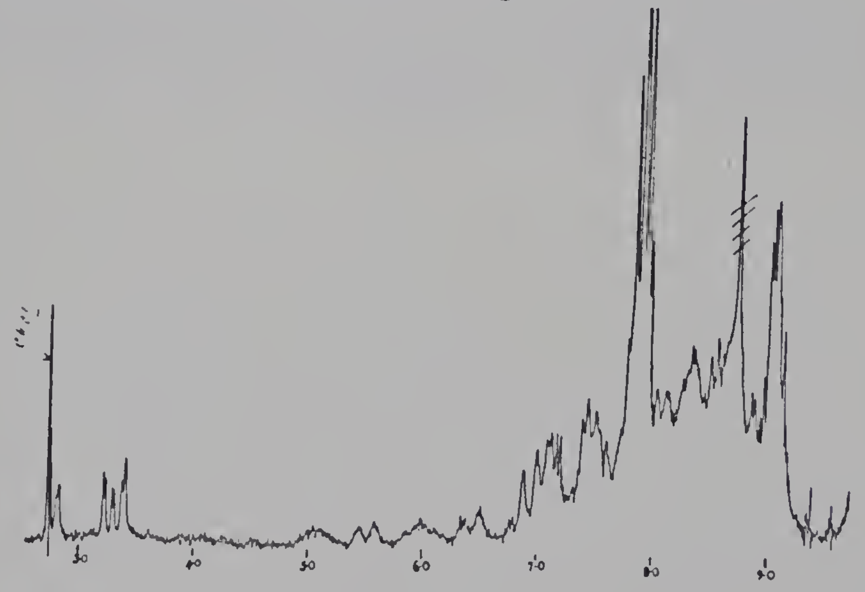


FIGURE 7. LYCOLUCINE ($CDCl_3$)

FIGURE 8. LUCIDINE-A.

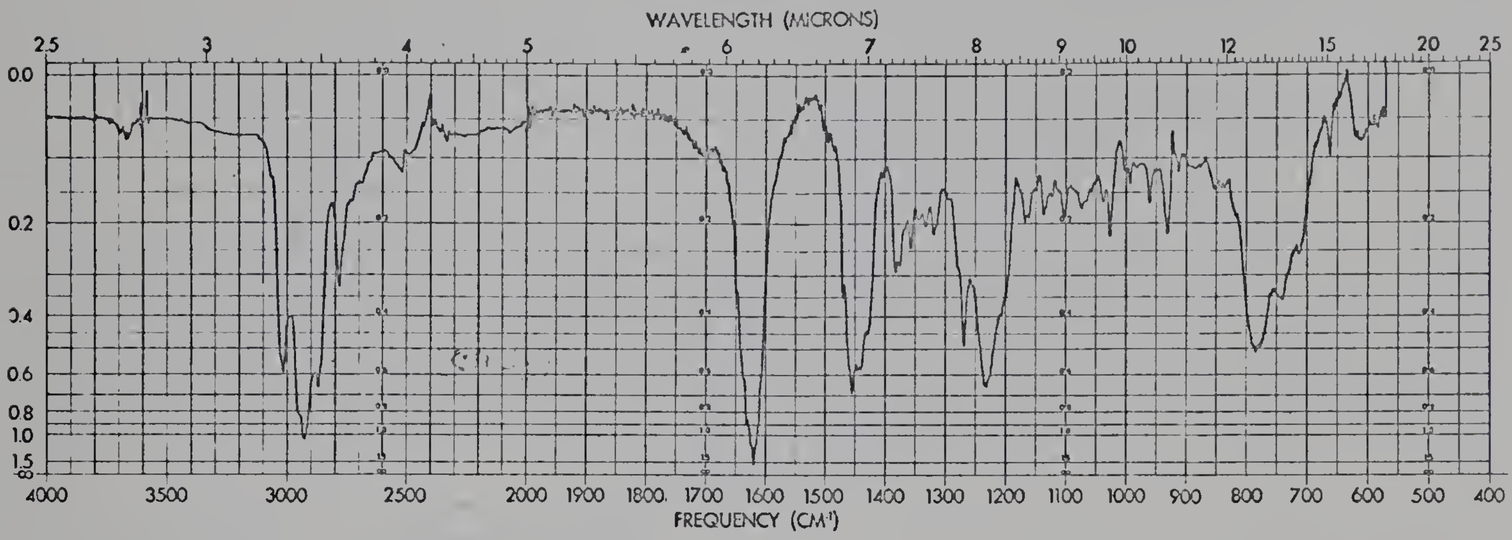
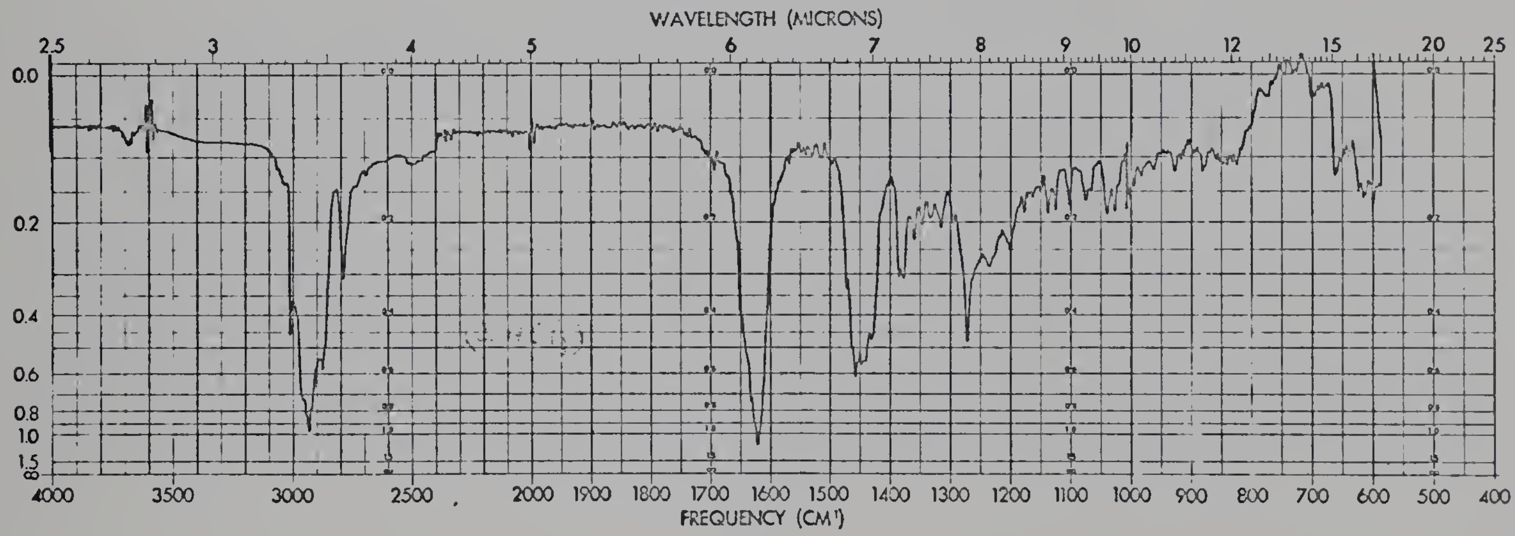


FIGURE 9. LUCIDINE-B.



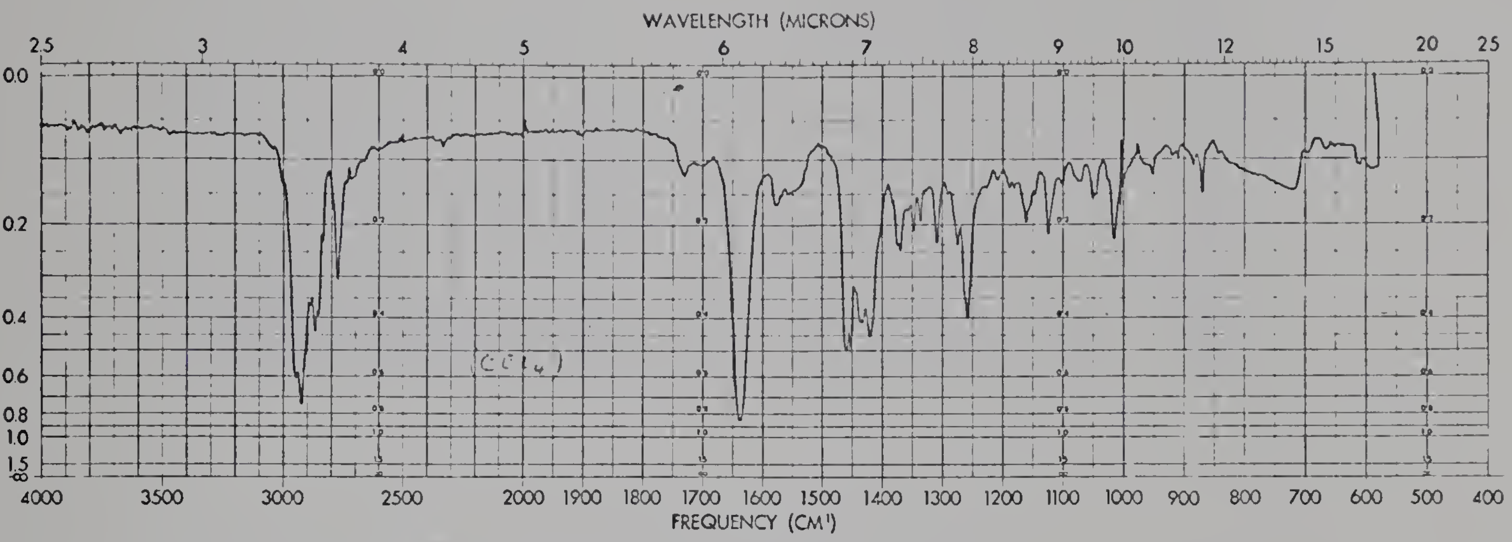


FIGURE 10. LYCOLUCINE

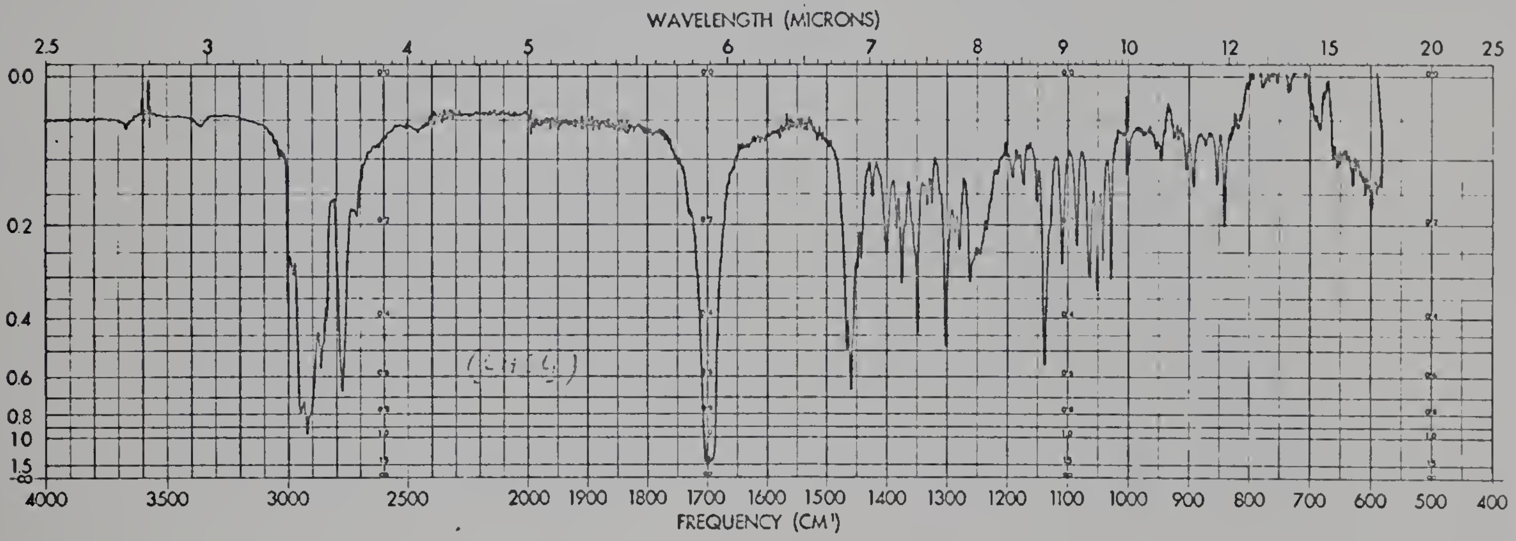


FIGURE 11. LUCIDULINE.

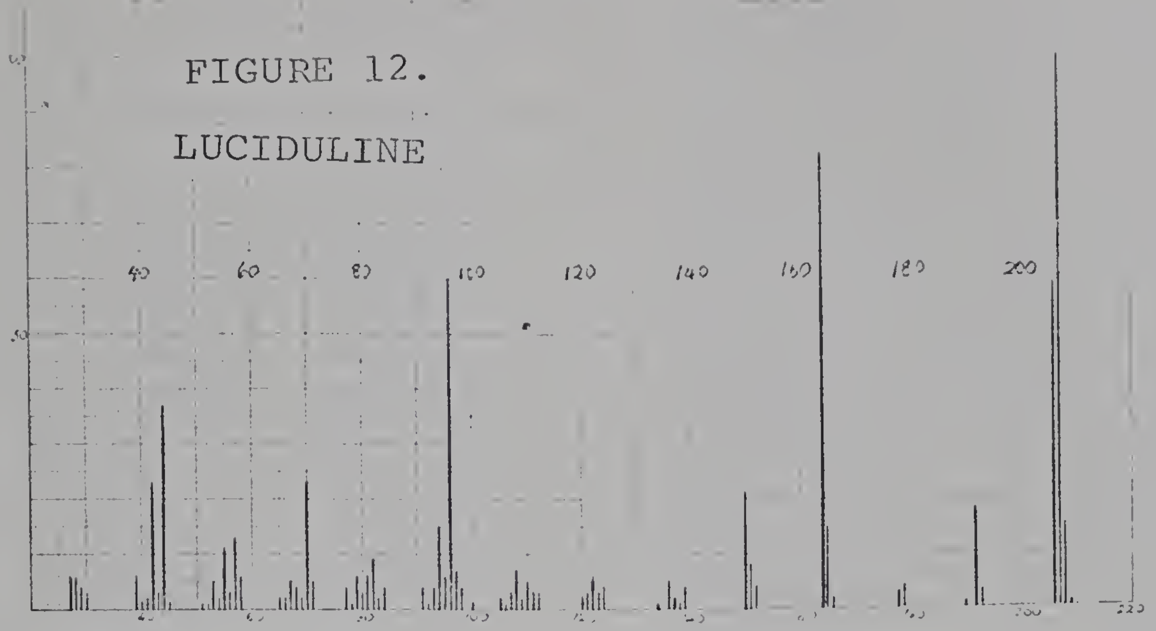


FIGURE 12.

LUCIDULINE

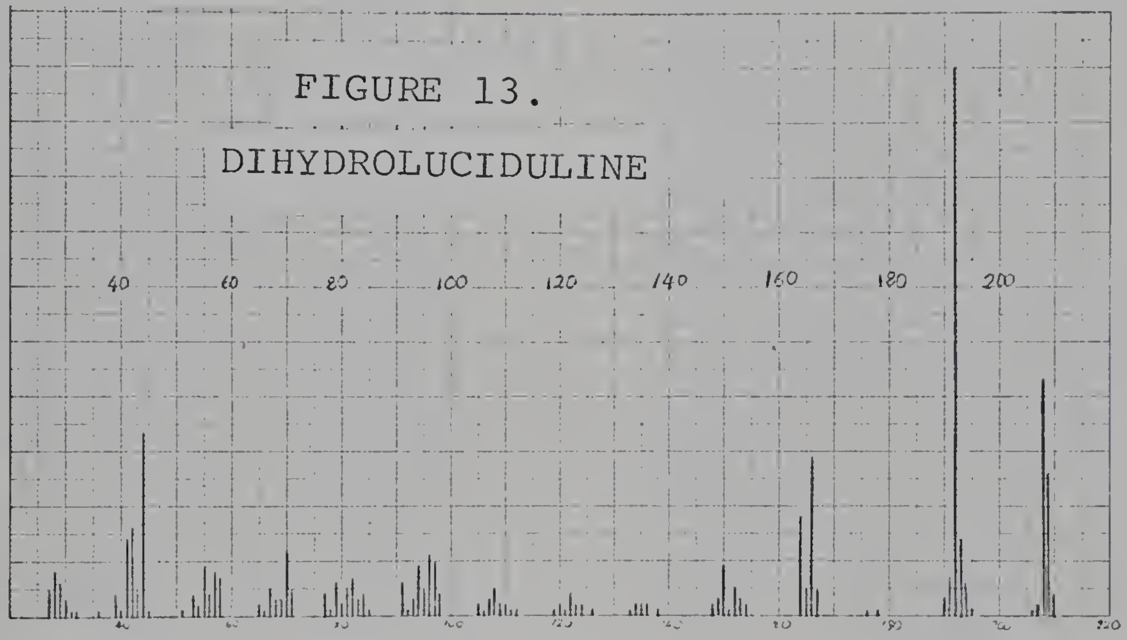


FIGURE 13.

DIHYDROLUCIDULINE

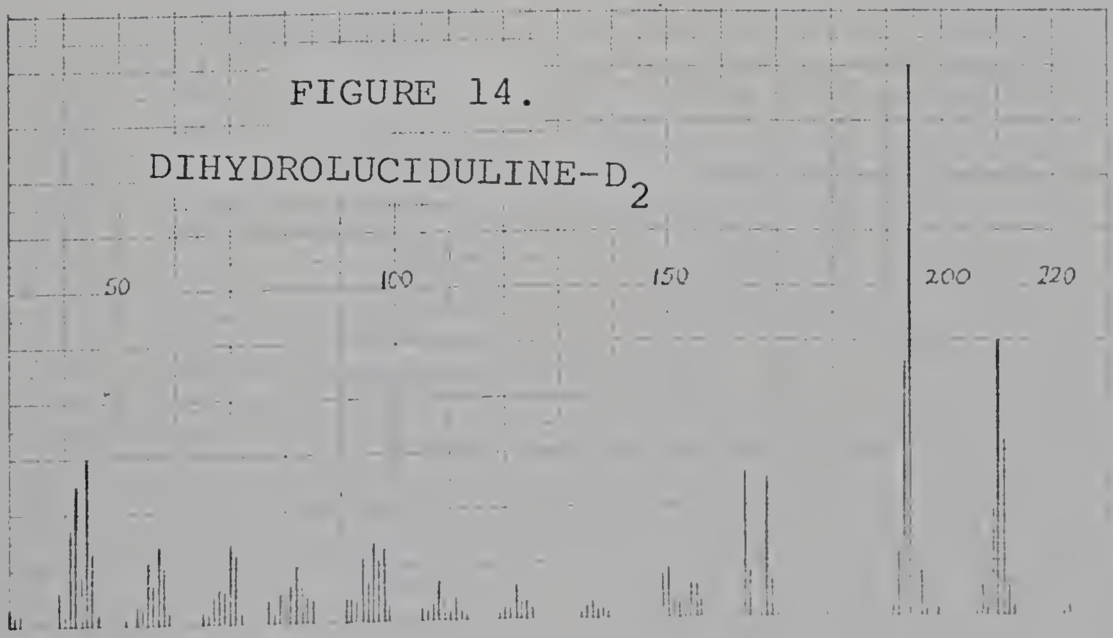


FIGURE 14.

DIHYDROLUCIDULINE-D_2

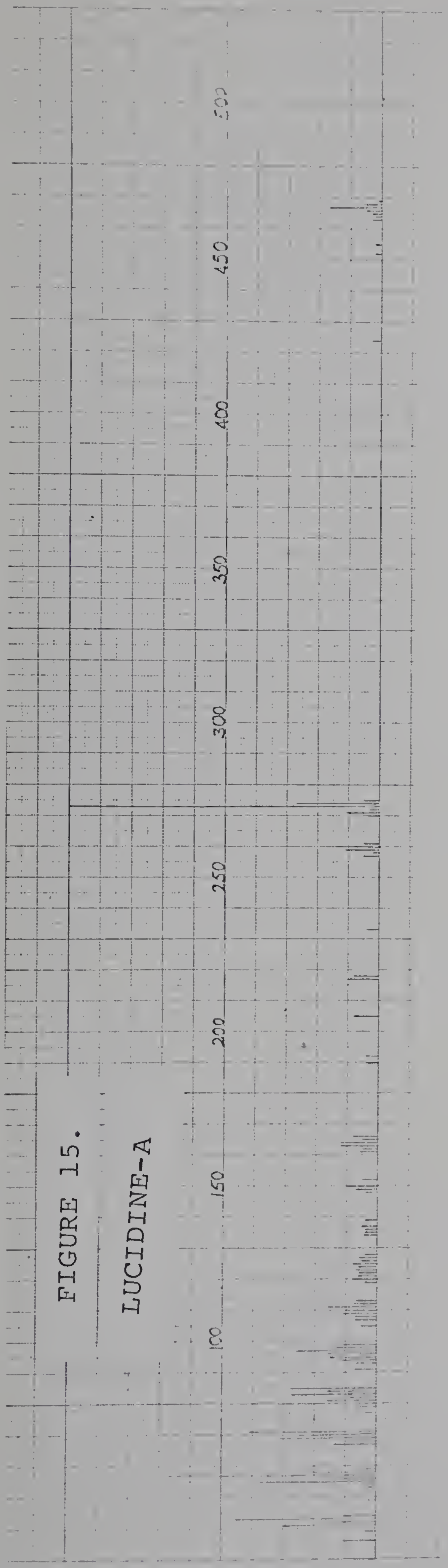


FIGURE 15.

LUCIDINE-A

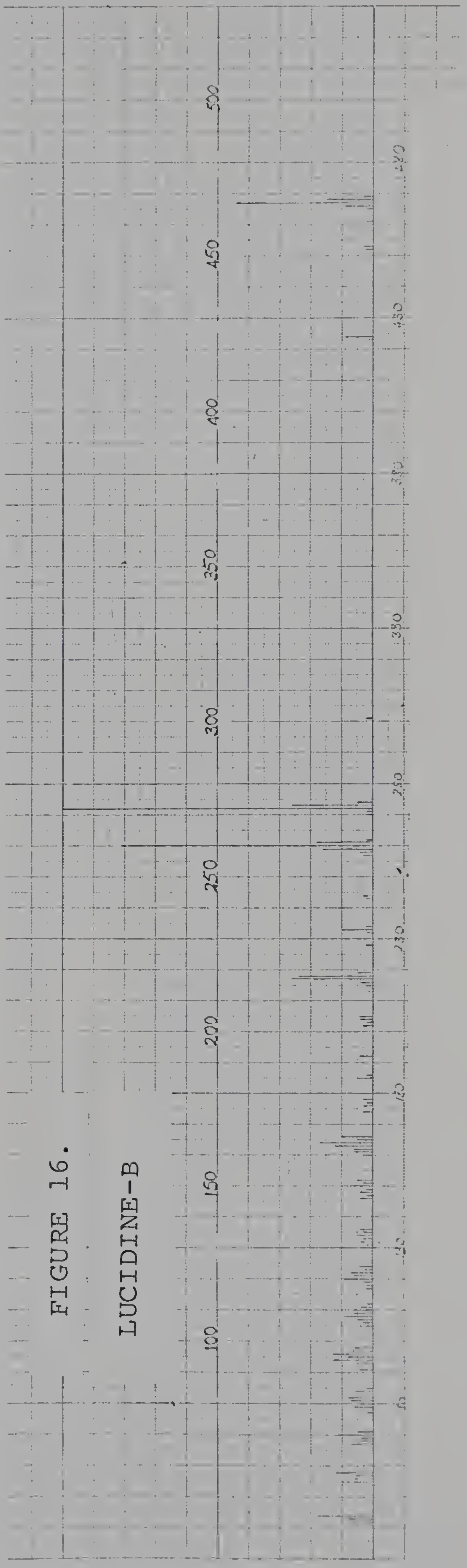


FIGURE 16.

LUCIDINE-B

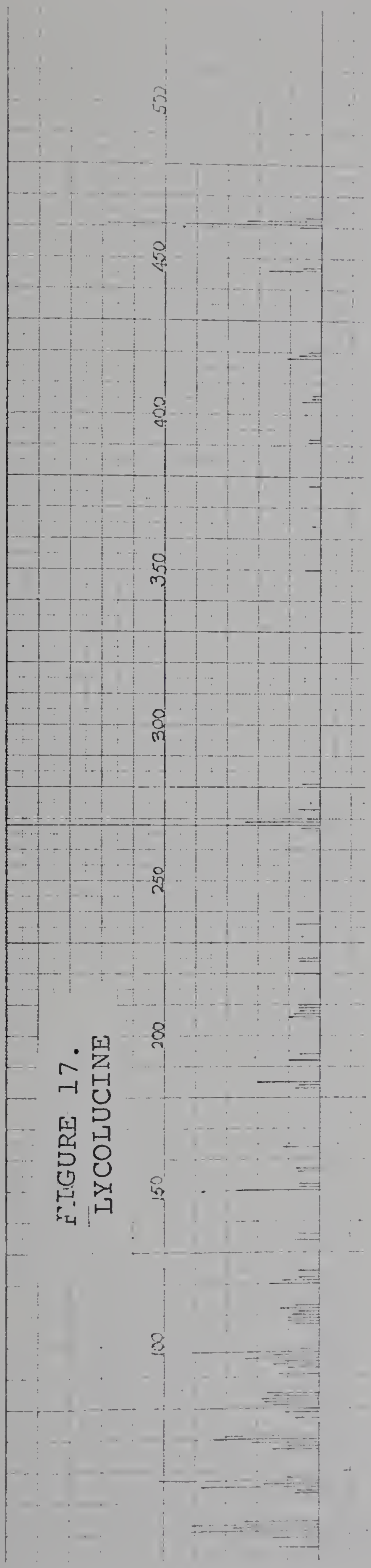


FIGURE 17.
LYCOLUCINE

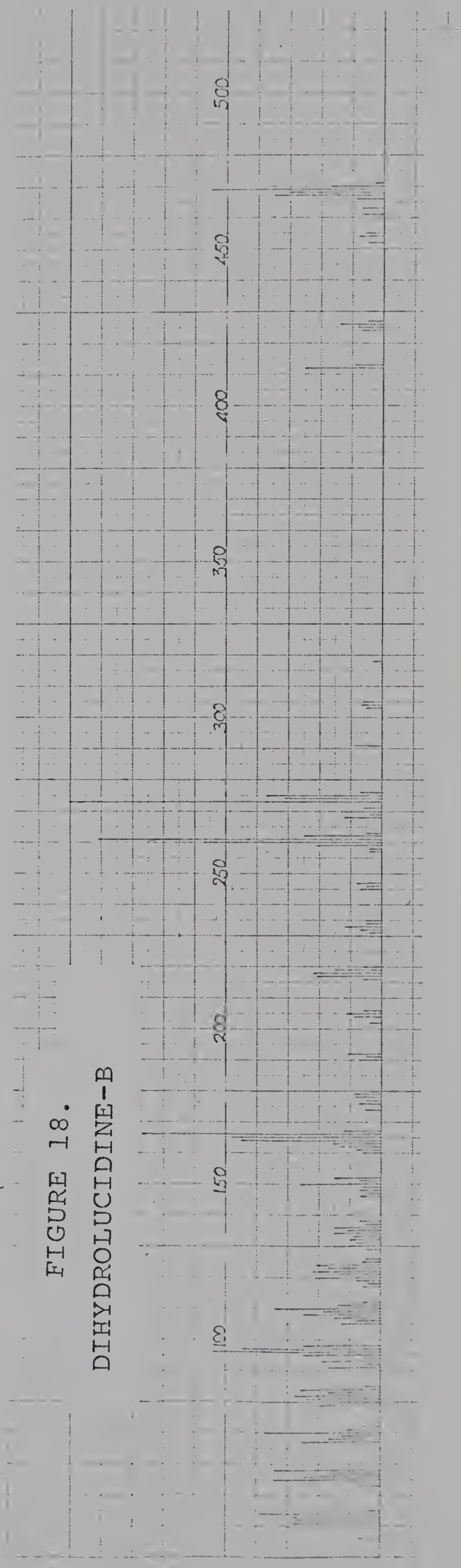


FIGURE 18.
DIHYDROLUCIDINE-B

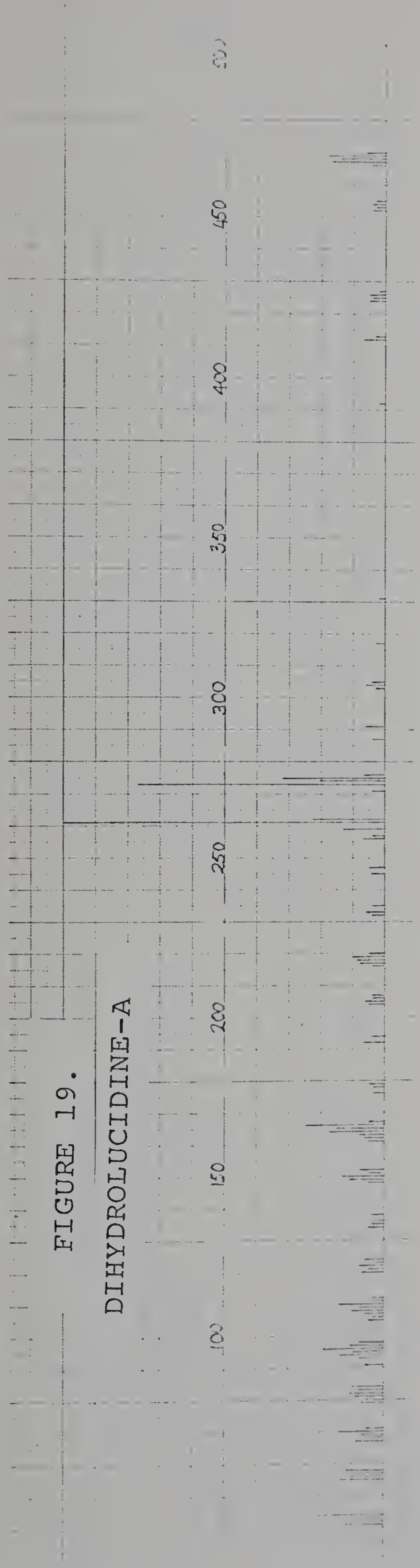


FIGURE 19.

DIHYDROLUCIDINE-A

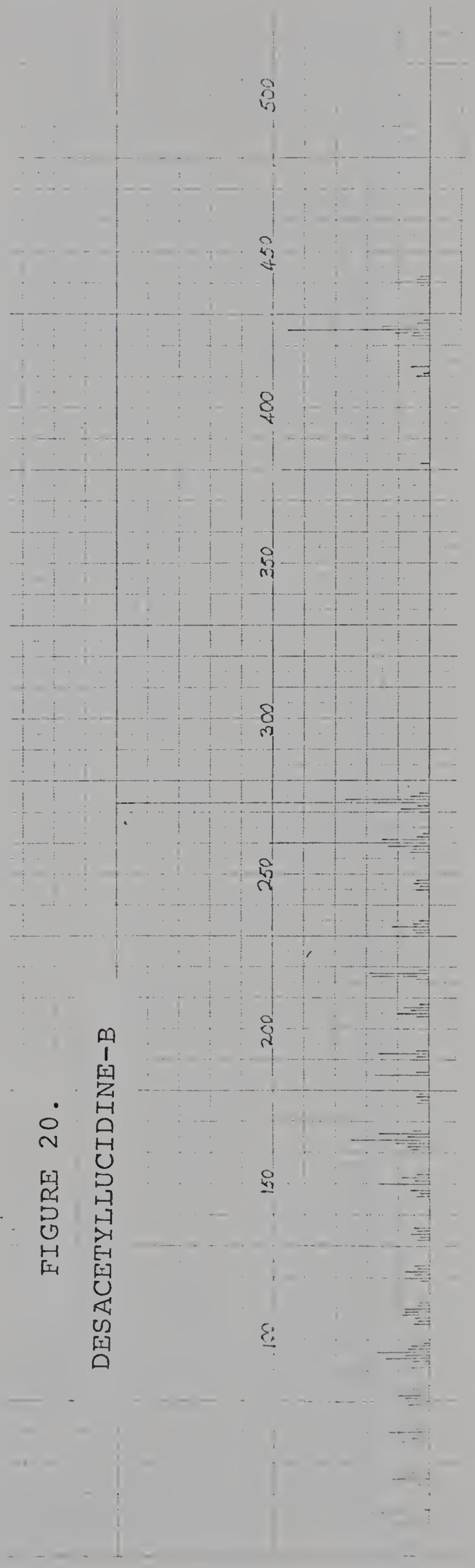


FIGURE 20.

DESACETYLLUCIDINE-B

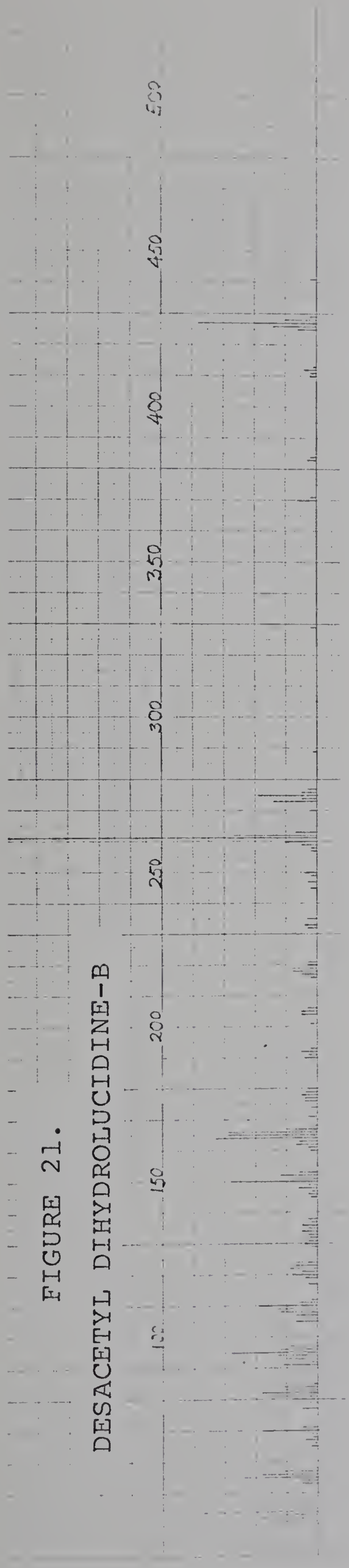


FIGURE 21.

DESACETYL DIHYDROLUCIDINE-B

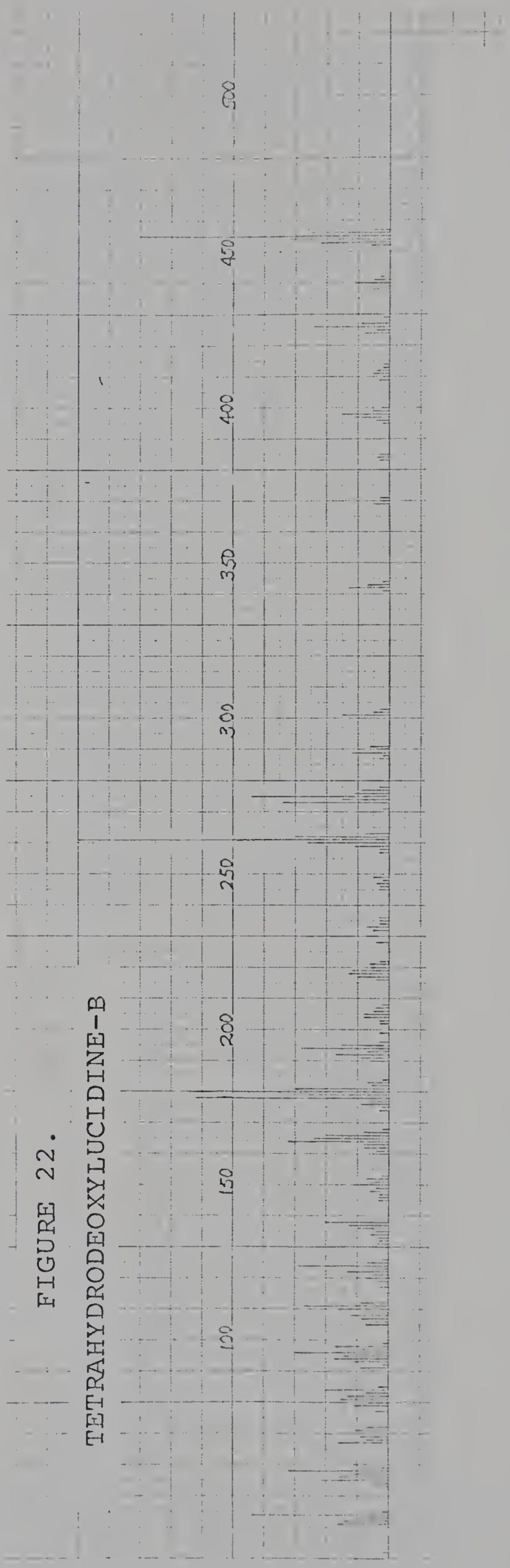


FIGURE 22.

TETRAHYDRODEOXYLUCIDINE-B

FIGURE 23.

TETRAHYDRODEOXYLUCIDINE-B-d_3

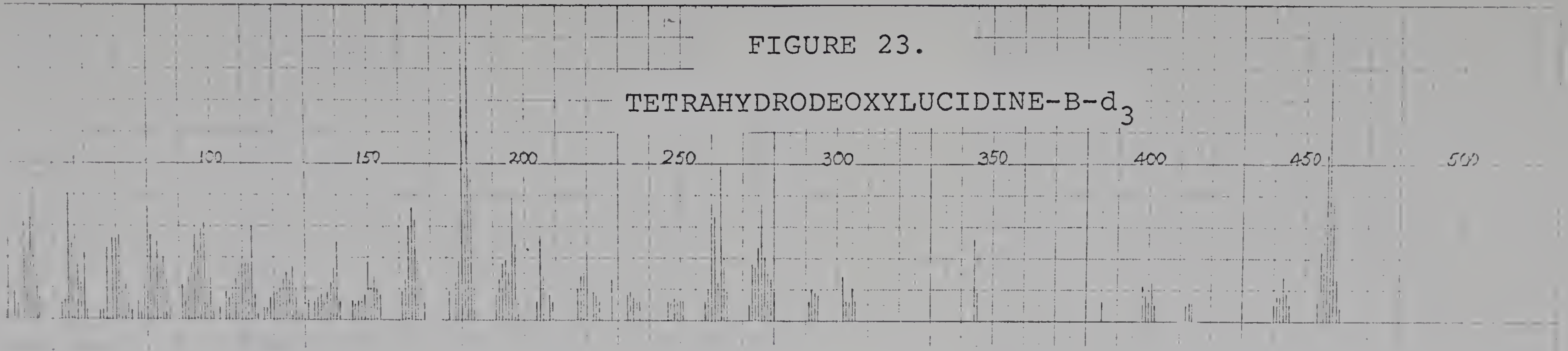
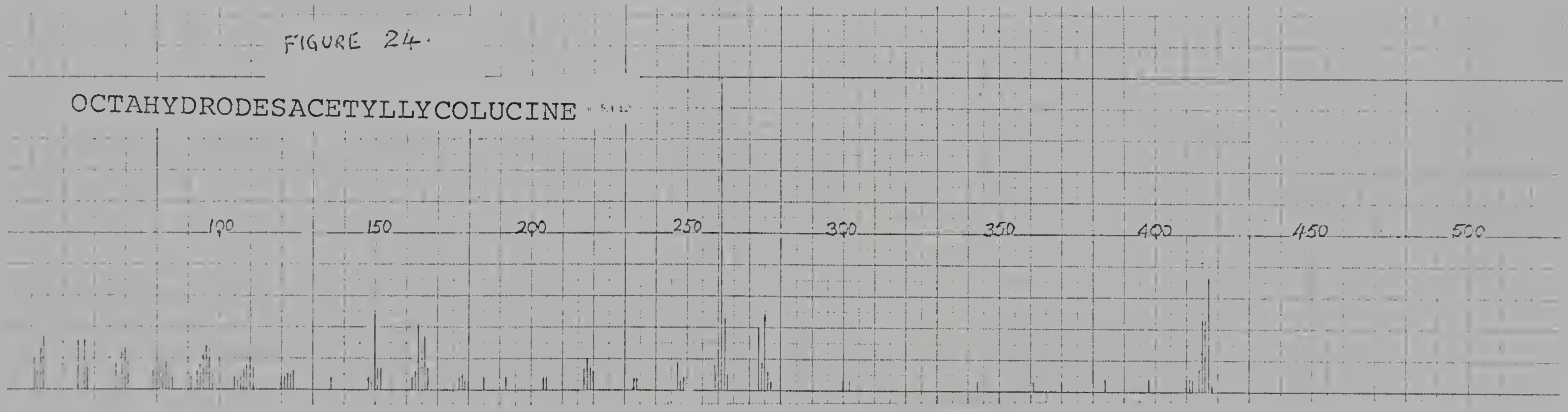


FIGURE 24.

OCTAHYDRODESACETYLLYCOLUCINE



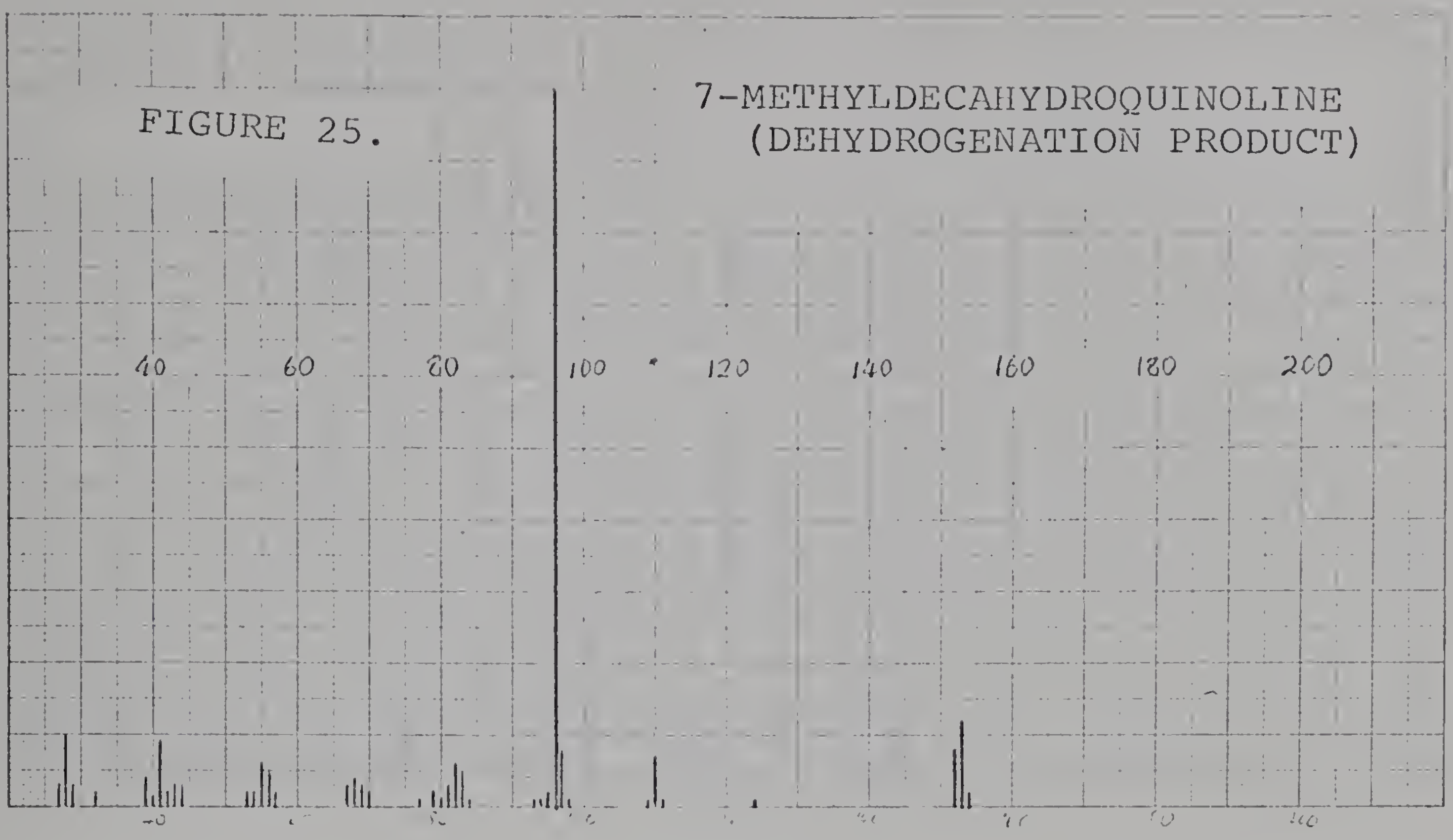


FIGURE 25.

7-METHYLDECAHYDROQUINOLINE (DEHYDROGENATION PRODUCT)

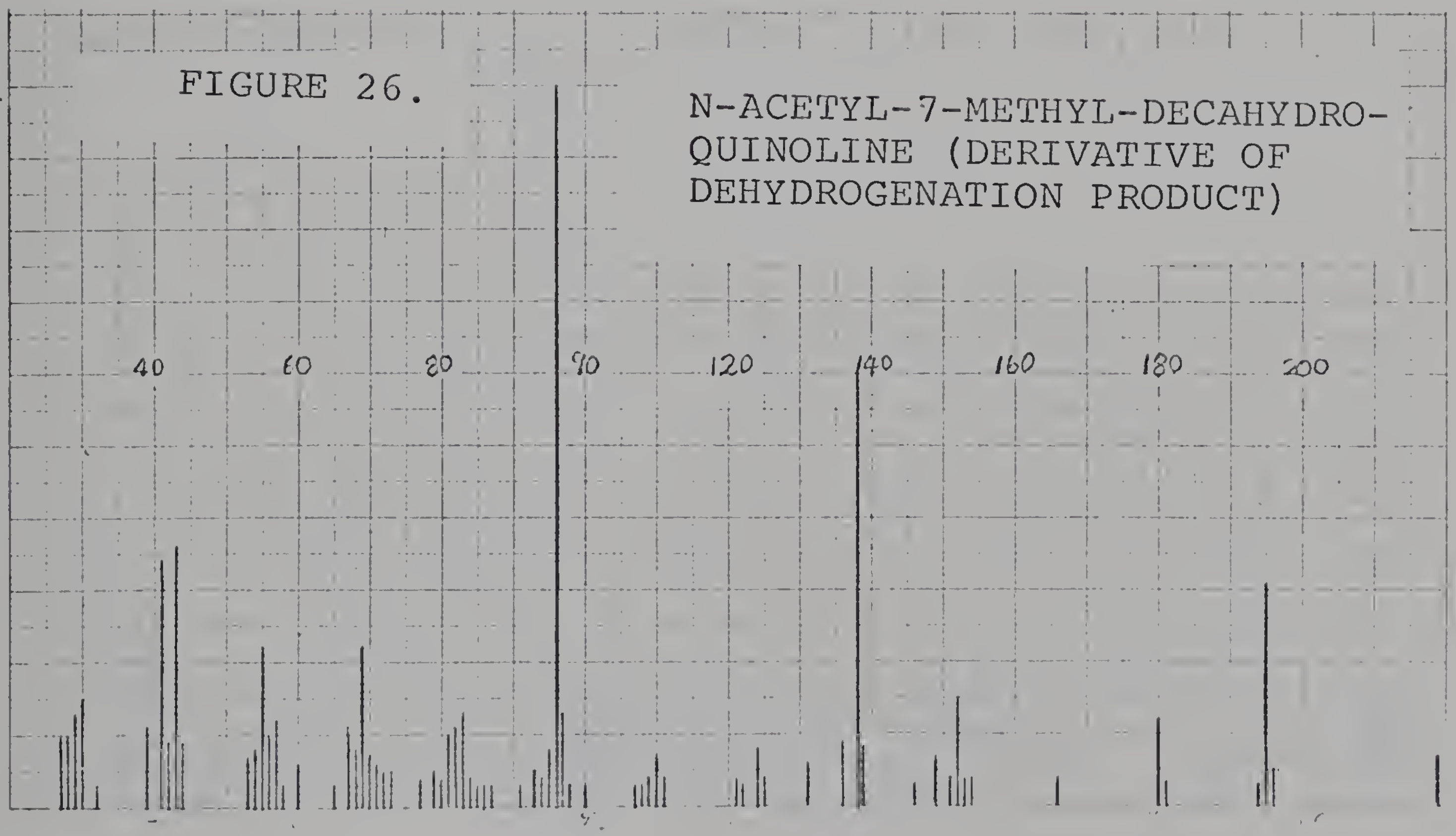


FIGURE 26.

N-ACETYL-7-METHYL-DECAHYDROQUINOLINE (DERIVATIVE OF DEHYDROGENATION PRODUCT)

FIGURE 27.

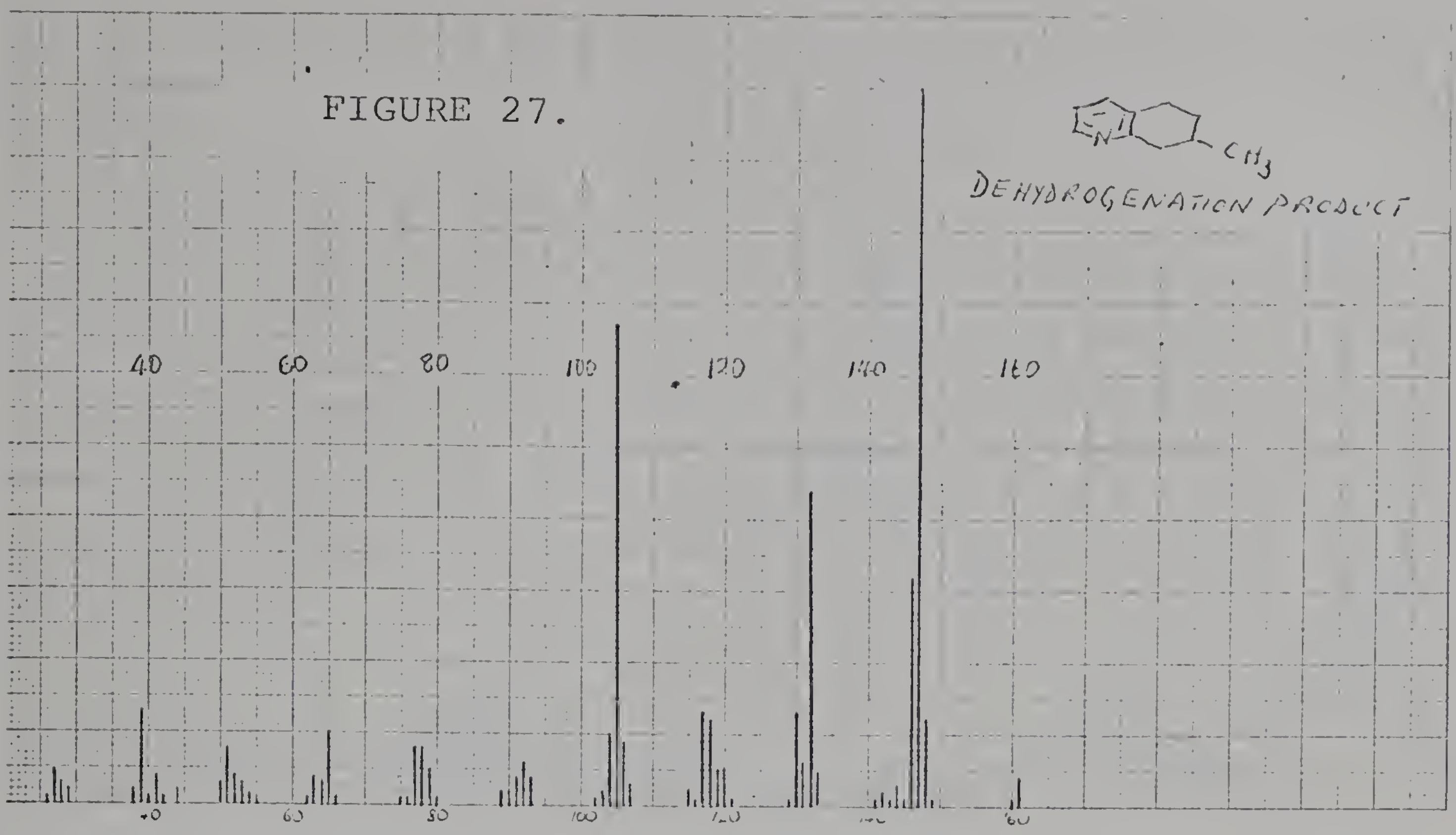
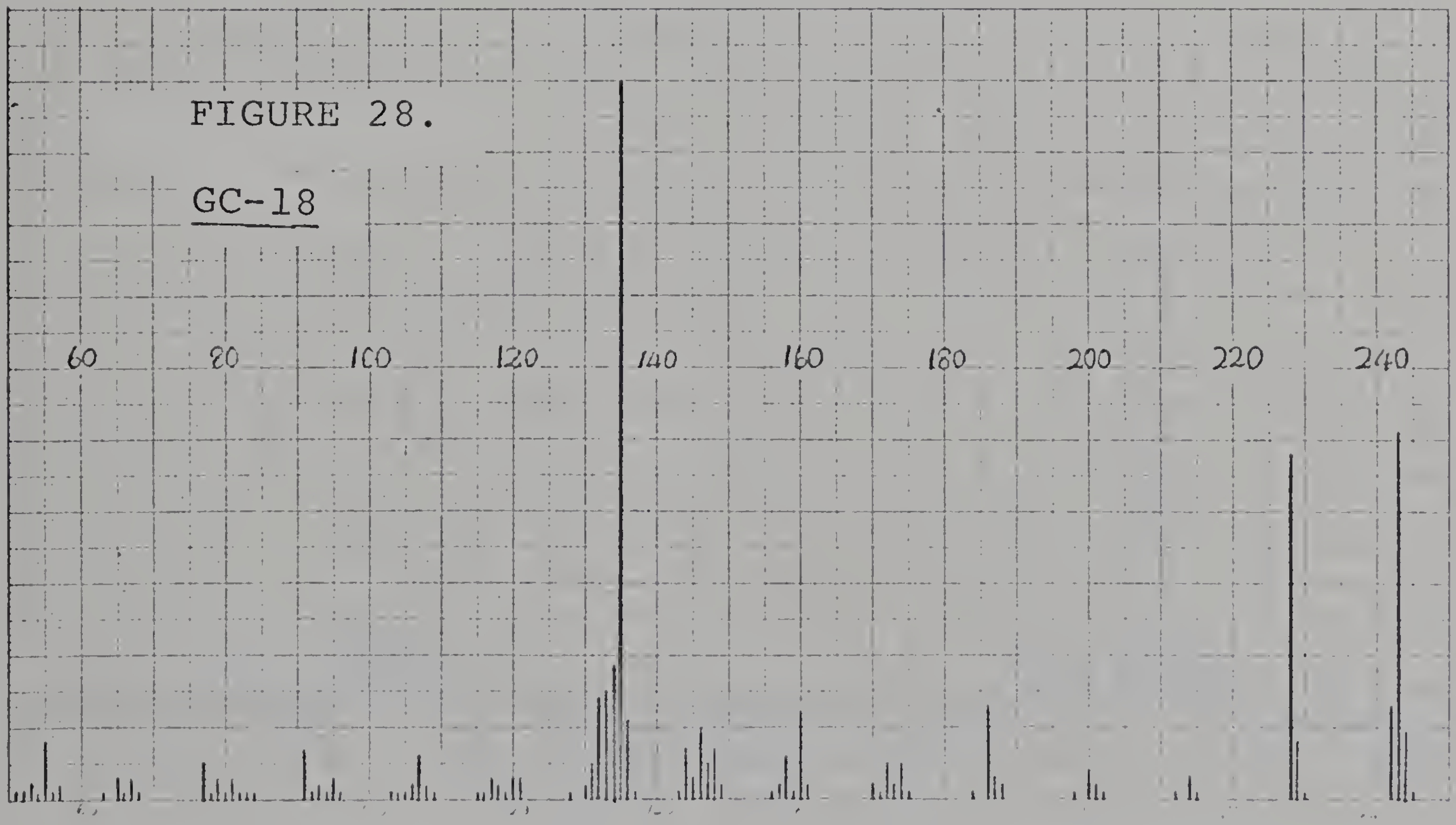


FIGURE 28.

GC-18



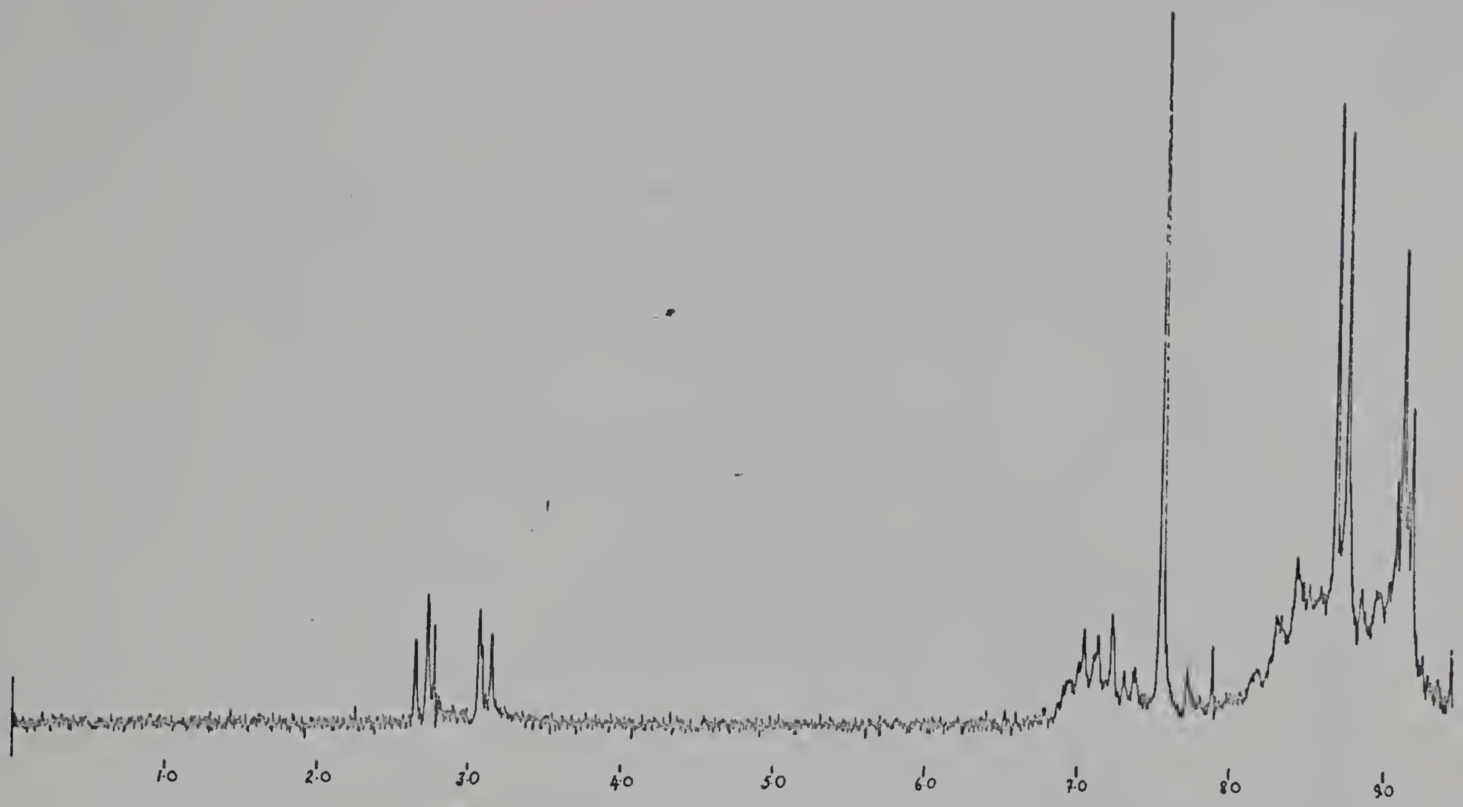


FIGURE 29. GC-18 ($CDCl_3$)

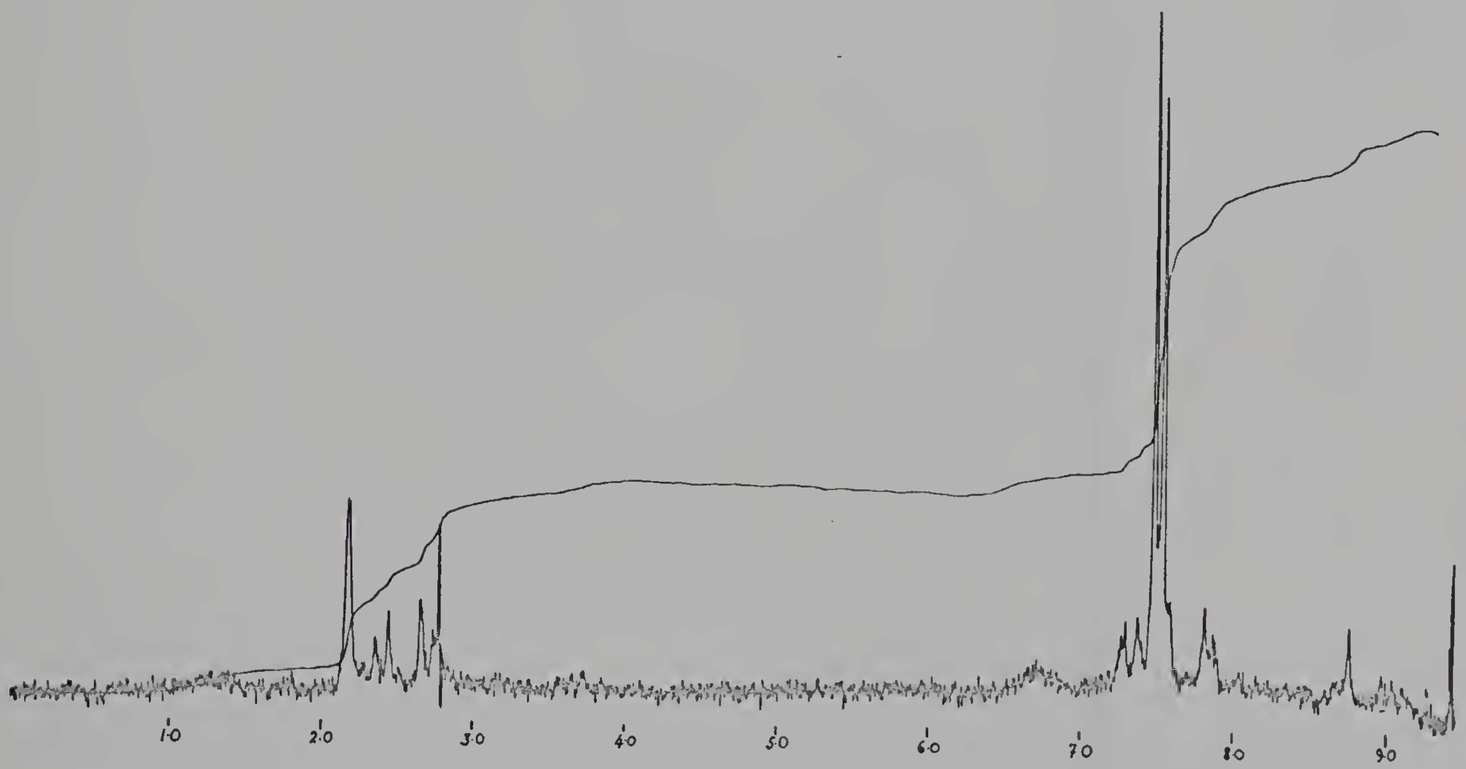


FIGURE 30. A DIMETHYLQUINOLINE ($CDCl_3$)

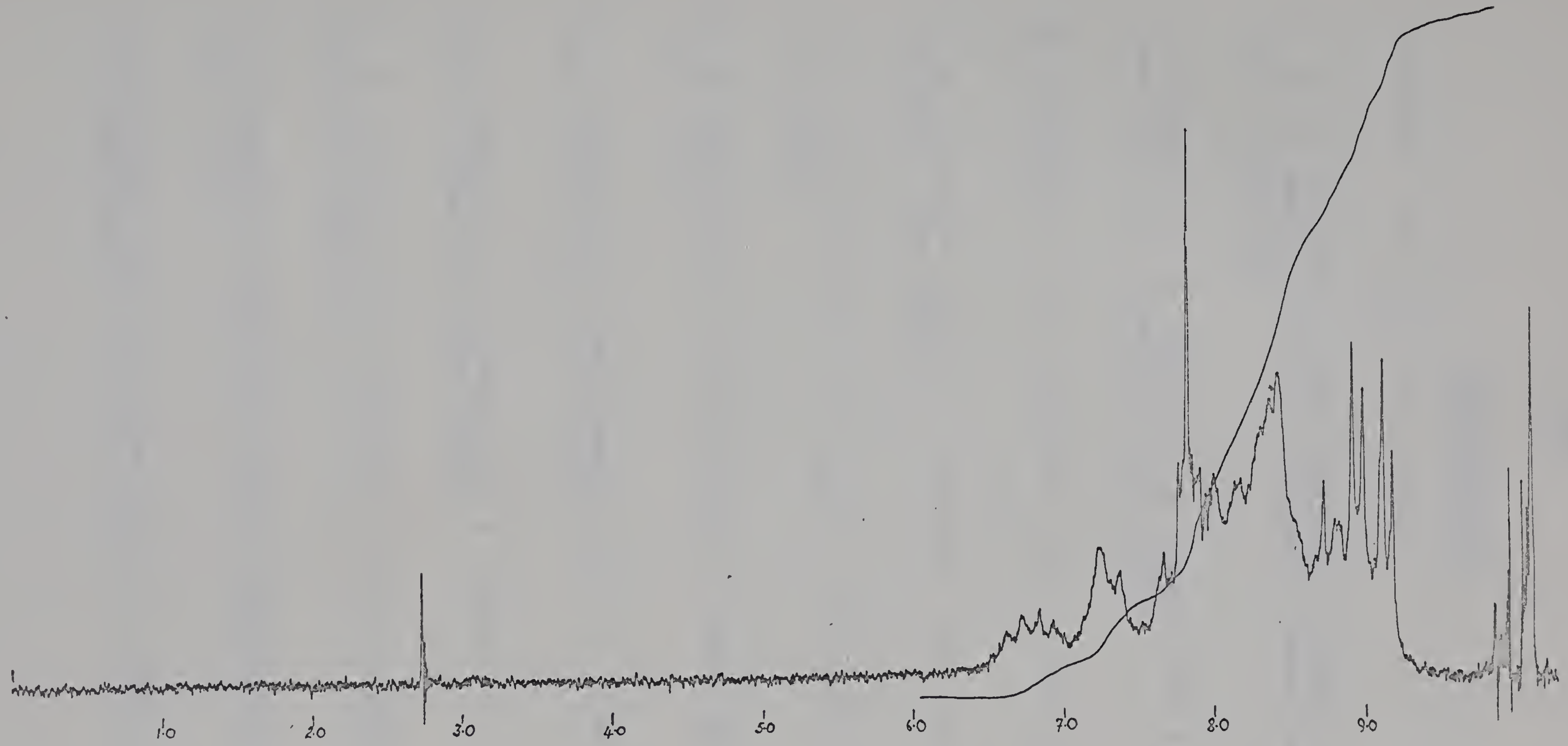


FIGURE 31. DESACETYLLUCIDINE-B ($CDCl_3$).

BIBLIOGRAPHY

1. K.Bödeker, Ann. 208, 363 (1881).

2. R. H. F. Manske in "The Alkaloids" Volume V, R. H. F. Manske, Editor, Academic Press, New York, 1955. p.295-299.

3. K. Wiesner, W. A. Ayer, L. R. Fowler and Z. Valenta, Chem. and Ind. (London), 564 (1957).

4. M. Przybylska and L. Marion, Can. J. Chem. 35, 1075 (1957).

5. W. A. Harrison and D. B. MacLean, Chem. and Ind. (London), 261 (1960).

6. R. H. Burnell and D. R. Taylor, Tetrahedron 15, 173 (1961).

7. W. A. Ayer and D. A. Law, Can. J. Chem. 40, 2088 (1962).

8. W. A. Ayer, J. A. Berezowsky and D. A. Law, Can. J. Chem. 41, 469 (1963).

9. W. A. Ayer, W. R. Bowman and P. Kebarle, Can. J. Chem. 43, 328 (1965).

10. W. A. Ayer and G. G. Iverach, Can. J. Chem. 42, 2514 (1964).

11. R. H. Burnell and D. R. Taylor, Tetrahedron 18, 1467 (1962).

12. F.A.L. Anet and N. H. Khan, Can. J. Chem. 37, 1589 (1959).

13. F.A.L. Anet, Tetrahedron Lett. 20, 13 (1960).

14. F.A.L. Anet and N. H. Khan, Chem. and Ind. (London), 1238 (1960).

15. W. N. French and D. B. MacLean, Can. J. Chem. 39, 2100 (1961).

16. B. Altenkirk, Private Communication. L23 is epi-lycodoline.

17. J.C.F. Young and D. B. MacLean, Can. J. Chem. 41, 2731 (1963).

18. W. A. Szarek, K. H. Adams, M. Curcumcelli-Rodostamo, and D. B. MacLean, Can. J. Chem. 42, 2584 (1964).

19. F.A.L. Anet, M. Z. Haq, N. H. Khan, W. A. Ayer, R. Hayatsu, S. Valverde-Lopez, P. Deslongchamps, W. Riess, M. Ternbah, Z. Valenta and K. Wiesner, Tetrahedron Letters, 751 (1964).

20. W. A. Ayer and G. G. Iverach, Can. J. Chem. 38, 1823 (1960).

21. W. A. Ayer, J. A. Berezowsky and G. G. Iverach, Tetrahedron 18, 567 (1962).

22. S. N. Alam, K.A.H. Adams and D. B. MacLean, Can. J. Chem. 42, 2456 (1964).

23. Z. Valenta, H. Yoshimura, E. F. Rogers, M. Ternbah, and K. Wiesner, Tetrahedron Lett. 10,26, (1960).

24. Y. Inubushi, H. Ishii, B. Yasui, M. Hashimoto and T. Harayama, Tetrahedron Letters, 1537 (1966).

25. B. Yasui, H. Ishii, T. Harayama, R. Nishino and Y. Inubushi, Tetrahedron Letters, 3967 (1966).

26. W. A. Ayer, T. E. Habgood, V. Deulofeu, and H. R. Juliani, Tetrahedron 21, 2169 (1965).

27. Y. Inubushi, H. Ishii and T. Harayama, Chem. and Pharm. Bull. (Japan) 15, 250 (1967).

28. H. Ishii, B. Yasui, T. Harayama and Y. Inubushi, Tetrahedron Letters, 6215 (1966).

29. Y. Inubushi, H. Ishii, T. Harayama, R. H. Burnell, W. A. Ayer and B. Altenkirk, Tetrahedron Letters, 1069 (1967).

30. R. H. Burnell, C. G. Chin, B. S. Mootoo and D. R. Taylor, Can. J. Chem. 41, 3091 (1963).

31. W. A. Ayer, J. K. Jenkins, S. Valverde-Lopez and R. H. Burnell, Tetrahedron Letters, 2201 (1964).

32. W. A. Ayer, J. K. Jenkins, S. Valverde-Lopez and R. H. Burnell, Can. J. Chem. 45, 433 (1967).

33. H. Conroy, Tetrahedron Lett. 10, 34, (1960).

34. W. A. Ayer and K. Piers, Can. J. Chem. 45, 451 (1967).

35. K. Wiesner, Fortschr. Chem. Org. Naturstoffe 20, 271-97 (1962). Springer-Verlag.

36. W. A. Ayer, D. A. Law and K. Piers, Tetrahedron Letters, 2959 (1964).

37. Y. Inubushi, H. Ishii, B. Yasui and T. Harayama, Tetrahedron Letters, 1551 (1966).

38. W. A. Ayer. Private Communication. See however, Reference 29.

39. Z. Valenta in "Technique of Organic Chemistry" A. Weissberger, Editor, Volume XI, Part I. Interscience, New York, 1963. pp.581-642.

40. J. F. King and P. de Mayo in "Molecular Rearrangements", P. de Mayo, Editor Vol.II. Interscience, New York, 1964 pp. 772-781.

41. E. W. Warnhoff in "Molecular Rearrangements", P. de Mayo, Editor. Vol. II. Interscience, New York, 1964. pp.879-907.

42. F. A. L. Anet and L. Marion, Can. J. Chem. 33, 849 (1955).

43. Z. Valenta, P. Deslongchamps, M. H. Raschid, R. H. Wightman and J. S. Wilson, Tetrahedron Letters, 1559 (1963).

44. T. L. Karle and J. Karle, Tetrahedron Letters, 2065 (1963).

45. Z. Valenta, P. Deslongchamps, M. H. Raschid, R. H. Wightman and J. S. Wilson, Can. J. Chem. 44, 2525 (1966).

46. J. A. Berezowsky, M.Sc. Thesis, University of Alberta, 1962, p.33.

47. J. A. Berezowsky. Unpublished work.

48. B. Loev and K. M. Snader. Chem. and Ind. (London), 15 (1965).

49. L. C. Craig and D. Craig in "Technique of Organic Chemistry" A. Weissberger, Editor, Volume III, Part I. Second Edition. Interscience, New York, 1956 pp.242-297.

50. A. I. Vogel, "A Textbook of Quantitative Inorganic Analysis", Second Edition. Longmans-Green (LOndon), p.868.

51. R. H. F. Manske and L. Marion, Can. J. Research 24B, 57 (1946).

52. R. N. Jones and C. Sandorfy in "Technique of Organic Chemistry", A. Weissberger, Editor, Volume IX. Interscience, New York, 1963 p.148.

53. J.C.N. Ma and E. W. Warnhoff, Can. J. Chem. 43, 1489 (1965).

54. S. Sternhell, Rev. Pure and Applied Chem. 14, 15 (1964).

55. D. B. MacLean, Can. J. Chem. 41, 2564 (1963).

56. H. Budzikiewicz, C. Djerassi and D. H. Williams, "Interpretation of Mass Spectra of Organic Compounds", Holden-Day, San Francisco, 1964, p.162.

57. W. Treibs, Ann. 576, 116 (1952).

58. C. H. Heathcock, R. A. Badger, and J. W. Patterson, Jr., J. Am. Chem. Soc., 89, 4133 (1967).

59. R. U. Lemieux and E. von Rudloff, Can. J. Chem. 33, 1701 (1955).

60. P. de Mayo in "Technique of Organic Chemistry" A. Weissberger, Editor, Vol.XI, Part II. Interscience, New York, 1963. p.1102.

61. (a) J. R. Hargreaves and P. W. Hickmott, Tetrahedron Letters, 4173 (1966).

(b) G. Eglington, J. Martin and W. Parker, J.Chem. Soc., 1243 (1965).

62. N. S. Bhacca and D. H. Williams, "Applications of NMR Spectroscopy in Organic Chemistry." Holden-Day, San Francisco, 1964. page 64.

63. (a) M. Uskoković, H. Bruderer, C. von Planta, T. Williams, and A. Brossi, J.Am. Chem. Soc. 86, 3364 (1964).

(b) A. K. Bosé in S. K. Freeman, Editor, "Interpretive Spectroscopy" Reinhold, New York, 1965. page 211.

64. H. P. Hamlow, S. Okuda and N. Nakagawa, Tetrahedron Letters, 2553 (1964).

65. S. Yamaguchi, S. Okuda and N. Nakagawa, Chem. and Pharm. Bull. (Japan) 11, 1465 (1963).

66. A. R. Katritzky, Record Chem. Progress 23, 223 (1962).

67. T. M. Moyneham, K. Schofield, R. A. Y. Jones, and A. R. Katritzky, J. Chem. Soc., 2653 (1962).

68. W. Moffit, R. B. Woodward, A. Moscowitz, W. Klyne and C. Djerassi, J. Am. Chem. Soc. 83, 4013 (1961).

69. W. A. Ayer and D. A. Law. Private communication.

70. H. Budzikiewicz, C. Djerassi and D. H. Williams, "Structure Elucidation of Natural Products by Mass Spectrometry" Volumes I and II. Holden-Day, San Francisco, 1964. General review.

71. W. M. Bryant III, A. L. Burlingane, H. O. House, C. G. Pitt and B. A. Tefertiller, J.Org. Chem. 31, 3120 (1966).

72. C. A. Grob and P. W. Schiess, Angewandte Chemie. International Edition, 6, 1 (1967).

73. K. Biemann, "Mass Spectrometry" McGraw-Hill, New York, 1962 pages 223-227.

74. J. Meinwald and R. A. Schneider, J. Am. Chem. Soc. 87, 5218 (1965).

75. R. J. L. Andon, J. D. Cox and E. F. G. Herington, Trans. Farad. Soc. 50, 918 (1954).

76. K. Nakanishi, "Infrared Absorption Spectroscopy" Holden-Day, San Francisco, 1962 page 20.

77. ibid. pages 45-47.

78. L. J. Bellamy, "The Infra-red Spectra of Complex Molecules," Metheus, London, 1959. pages 267-271.

79. E. M. Wilson, Chem. and Ind. (London), 472 (1965).

80. J. F. King in "Technique of Organic Chemistry," A. Weissberger, Editor, Volume XI, Part I. Interscience, New York, 1963. page 375.

81. R. Bonnet, J. Chem. Soc., 2313 (1965).

82. D. A. Nelson and J. J. Worman, Tetrahedron Letters, 507 (1966).

83. F. F. Blicke in "Organic Reactions" Volume I. John Wiley, New York, 1942. pages 303-341.

84. S. W. Pelletier and P. C. Parthasarathy, J. Am. Chem. Soc. 87, 777 (1965).

85. (a) F. A. L. Anet, Can. J. Chem. 36, 902 (1958).

(b) G. Büchi, I. M. Goldman and D. W. Mayo, J. Am. Chem. Soc. 88, 3109 (1966).

86. "High Resolution NMR Spectra Catalog," Varian Associates, Palo Alto, California. Spectra #154 and 155.

87. D. McMinn. Private communication. Commercial 7-methylquinoline contains some 5-methylquinoline. Pure 7-methylquinoline was obtained via picrate formation.

88. D. McMinn. Private communication. 7-methylquinoline was reduced catalytically over Raney Nickel.

89. D. W. Mathieson, Editor, "Interpretation of Organic Spectra", Academic Press, London, 1965, page 53.

90. K. Biemann, "Mass Spectromety," McGraw-Hill, New York, 1962. page 130.

91. A. M. Duffield, H. Budzikiewicz, D. H. Williams and C. Djerassi, J. Am. Chem. Soc. 87, 810 (1965).

92. (a) F. Weiss, A. Isard and G. Bonnard, Bull. Soc. Chim. France, 2332 (1965).

(b) R. L. Levy, J. Gas Chromatography, 107 (1967).

93. J. M. Bobbitt "Thin-Layer Chromatography" Reinhold, New York, 1963. page 93.

94. A. I. Vogel, "A Textbook of Practical Organic Chemistry". Longmans, London, 1961. pages 262 and 792.

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